09/856336
AU 1645

=> s plasmid?
L1 327331 PLASMID?

=> s butanol?
L2 72022 BUTANOL?

=> s chaotrop?
L3 4823 CHAOTROP?

=> s l1 and l2 and l3
L4 6 L1 AND L2 AND L3

=> dup rem l4
PROCESSING COMPLETED FOR L4
L5 4 DUP REM L4 (2 DUPLICATES REMOVED)

=> d l5 ibib abs 1-4

L5 ANSWER 1 OF 4 WPIDS COPYRIGHT 2002 DERWENT INFORMATION LTD DUPLICATE 1
ACCESSION NUMBER: 2000-387779 [33] WPIDS
DOC. NO. CPI: C2000-117783
TITLE: Extraction of ***plasmid*** DNA from DNA-containing material using an organic solvent, a ***chaotrope*** and water, useful particularly for obtaining ***plasmid*** DNA from bacterial culture.
DERWENT CLASS: B04 D16
INVENTOR(S): BUTT, N J; JONES, C P; JOENES, C P
PATENT ASSIGNEE(S): (CAMB-N) CAMBRIDGE MOLECULAR TECHNOLOGIES LTD; (WHAT-N) WHATMAN BIOSCIENCE LTD
COUNTRY COUNT: 89
PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
WO 2000029563	A1	20000525	(200033)*	EN	16

RW: AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW NL
OA PT SD SE SL SZ TZ UG ZW
W: AE AL AM AT AU AZ BA BB BG BR BY CA CH CN CU CZ DE DK EE ES FI GB
GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC LK LR LS LT LU
LV MA MD MG MK MN MW MX NO NZ PL PT RO RU SD SE SG SI SK SL TJ TM
TR TT TZ UA UG US UZ VN YU ZA ZW
GB 2346615 A 20000816 (200040)
AU 2000011699 A 20000605 (200042)
EP 1131420 A1 20010912 (200155) EN
R: AL AT BE CH CY DE DK ES FI FR GB GR IE IT LI LT LU LV MC MK NL PT
RO SE SI

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
WO 2000029563	A1	WO 1999-GB3830	19991117
GB 2346615	A	GB 1998-25215	19981117
AU 2000011699	A	AU 2000-11699	19991117
EP 1131420	A1	EP 1999-972256	19991117
		WO 1999-GB3830	19991117

FILING DETAILS:

PATENT NO	KIND	PATENT NO
AU 2000011699	A Based on	WO 200029563
EP 1131420	A1 Based on	WO 200029563

PRIORITY APPLN. INFO: GB 1998-25215 19981117
AN 2000-387779 [33] WPIDS
AB WO 200029563 A UPAB: 20010410
NOVELTY - A novel extraction mixture for selectively extracting ***plasmid*** DNA from a DNA -containing material, comprises a water-immiscible organic solvent capable of supporting ***plasmid*** DNA, a ***chaotrope*** and water.
DETAILED DESCRIPTION - A novel method for isolating ***plasmid***
DNA from a DNA containing material which comprises ***plasmid*** DNA
and genomic DNA comprises:
(i) extracting the ***plasmid*** DNA into a water-immiscible organic solvent capable of supporting ***plasmid*** DNA, by mixing the material with the organic solvent, a ***chaotrope*** and water under conditions to denature the genomic DNA; and
(ii) recovering the ***plasmid*** DNA from the organic phase.
An INDEPENDENT CLAIM is also included for an extraction mixture for selectively extracting ***plasmid*** DNA from a water-immiscible organic solvent capable of supporting ***plasmid*** DNA, a ***chaotrope*** and water.
USE - The method can be used for the extraction of ***plasmid*** DNA from DNA-containing material such as a bacterial culture which may be lysed or unlysed.
ADVANTAGE - The method is capable of extracting ***plasmid*** DNA to high purity and with particularly low or zero contamination from genomic DNA.
Dwg.0/0

L5 ANSWER 2 OF 4 WPIDS COPYRIGHT 2002 DERWENT INFORMATION LTD
ACCESSION NUMBER: 2001-061875 [07] WPIDS
DOC. NO. CPI: C2001-017279
TITLE: Isolating extrachromosomal nucleic acids, by homogenizing sample, removing chromosomal nucleic acids, contacting sample with ***chaotropic*** agent to precipitate nucleic acids and recovering them.
DERWENT CLASS: A96 B04 D16
INVENTOR(S): DEDMAN, J R; KAETZEL, M A; REED, T D
PATENT ASSIGNEE(S): (UYCI-N) UNIV CINCINNATI
COUNTRY COUNT: 93
PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
WO 2000077235	A1	20001221	(200107)*	EN	44

RW: AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW MZ
NL OA PT SD SE SL SZ TZ UG ZW
W: AE AG AL AM AT AU AZ BA BB BG BR BY CA CH CN CR CU CZ DE DK DM DZ
EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC LK
LR LS LT LU LV MA MD MG MK MN MW MX NO NZ PL PT RO RU SD SE SG SI
SK SL TJ TM TR TT TZ UA UG US UZ VN YU ZA ZW
AU 2000056206 A 20010102 (200121)

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
WO 2000077235	A1	WO 2000-US16712	20000616
AU 2000056206	A	AU 2000-56206	20000616

FILING DETAILS:

PATENT NO	KIND	PATENT NO
AU 2000056206	A Based on	WO 200077235

PRIORITY APPLN. INFO: US 1999-139423P 19990616
AN 2001-061875 [07] WPIDS
AB WO 200077235 A UPAB: 20011129
NOVELTY - A new method for isolating extrachromosomal nucleic acids (NAs), comprises homogenizing a sample, removing chromosomal nucleic acids, contacting the sample with a ***chaotropic*** agent to precipitate nucleic acids and recovering nucleic acids
DETAILED DESCRIPTION - A novel method of isolating

extrachromasomal
NAs in a purified form comprises:
(a) homogenizing a biological sample;
(b) removing all of the chromosomal NAs from the biological sample;
(c) contacting the biological sample containing extrachromosomal NAs with a ***chaotropic*** solution under conditions which permit the NAs
in the supernatants to precipitate; and
(d) recovering the extrachromosomal NAs.
An INDEPENDENT CLAIM is also included for a method of isolating NAs
in a purified form comprising:
(i) homogenizing a biological sample containing NAs of interest;
(ii) partially purifying the NAs from the biological sample by adhering the NAs of interest to a retaining device;
(iii) contacting a gel containing the sample with a ***chaotropic*** solution under conditions which permit the NAs to precipitate; and
(iv) recovering the NAs.
USE - The method can be used for separating extracellular NAs from e.g. recombinant bacteriophage, ***plasmid*** , cosmid, yeast expression vector, viral expression vector, or retrovirus (claimed). The method can be used for isolating NAs in a cell system such as bacteria, yeast, insect, plant and mammalian systems (claimed).
ADVANTAGE - The methods can produce high yields of optimally pure
intact extrachromosomal DNA without the need for toxic chemicals or DNA-binding materials. The process can be completed in 2-3 hours.
Dwg.0/0

L5 ANSWER 3 OF 4 WPIDS COPYRIGHT 2002 DERWENT INFORMATION LTD DUPLICATE 2
ACCESSION NUMBER: 2000-194941 [17] WPIDS
DOC. NO. CPI: C2000-060362
TITLE: Isolating circular nucleic acids, particularly ***plasmids*** , from a mixture of nucleic acids comprises treating the mixture under alkaline conditions with a solid matrix in the presence of a ***chaotrophic*** substance.
DERWENT CLASS: B04 D16
INVENTOR(S): KANG, J; SAUER, P
PATENT ASSIGNEE(S): (QIAG-N) QIAGEN GMBH
COUNTRY COUNT: 27
PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
WO 9961603	A1	19991202	(200017)*	EN	32

RW: AT BE CH CY DE DK ES FI FR GB GR IE IT LU MC NL PT SE
W: JP US
EP 969090 A1 20000105 (200017) EN
R: AL AT BE CH CY DE DK ES FI FR GB GR IE IT LI LT LU LV MC MK NL PT
RO SE SI
EP 1088064 A1 20010404 (200120) EN
R: AT BE CH CY DE DK ES FI FR GB GR IE IT LI LU MC NL PT SE

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
WO 9961603	A1	WO 1999-EP3660	19990527
EP 969090	A1	EP 1998-109593	19980527
EP 1088064	A1	EP 1999-938037	19990527
		WO 1999-EP3660	19990527

FILING DETAILS:

PATENT NO	KIND	PATENT NO
EP 1088064	A1 Based on	WO 9961603

PRIORITY APPLN. INFO: EP 1998-109593 19980527
AN 2000-194941 [17] WPIDS
AB WO 9961603 A UPAB: 20000405
NOVELTY - Separating and/or isolating circular nucleic acids from a mixture having different species of nucleic acids other than circular nucleic acids, comprises treating the mixture under alkaline conditions at a pH more than 8 with a solid matrix consisting essentially of a silica material in presence of at least one ***chaotropic*** substance.
DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for:
(1) an aqueous buffer comprising 6-9 M sodium thiocyanate, 0-20 vol.-% 1-4C-alcohols such as ethanol or isopropanol, 25-130 mM buffer substance preferably omega -amino acids; and
(2) a kit comprising the aqueous buffer of (1) and auxiliary materials such as columns with our without silicaceous material, suspensions of silicaceous material, additional buffers such as resuspension buffers, lysis buffers, washing buffers, elution buffers, instruction manual.
USE - For isolating and/or separating circular nucleic acids, particularly ***plasmids*** , from a mixture having different species of nucleic acids. Isolation of ***plasmid*** DNA is often a prerequisite for subsequent molecular biological experiments, e.g. PCR reactions, sequencing reactions, cloning reactions, restriction hydrolyses, transformations and transfections.
ADVANTAGE - Unlike all known methods for the isolation of ***plasmid*** DNA, the nucleic acids are isolated from bacterial crude lyzate avoiding the need to form cleared lyzate. (Clearing the lyzate is the most time-consuming step (e.g. by centrifugation) in ***plasmid*** isolation protocol or the major cost factor (e.g. by filtration)). The method is therefore simple and rapid.
Dwg.0/0

L5 ANSWER 4 OF 4 HCAPLUS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 1995:623433 HCAPLUS
DOCUMENT NUMBER: 123:27196
TITLE: Refolding of improperly folded polypeptides like recombinant insulin-like growth factor recovered from inclusion bodies
INVENTOR(S): Builder, Stuart; Hart, Roger; Lester, Phillip; Reifsnyder, David
PATENT ASSIGNEE(S): Genetech, Inc., USA
SOURCE: PCT Int. Appl., 74 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9506064	A1	19950302	WO 1994-US9120	19940815

W: CA, JP, US
RW: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE
US 5663304 A 19970902 US 1993-110664 19930820
CA 2168552 AA 19950302 CA 1994-2168552 19940815
EP 714406 A1 19960605 EP 1994-927917 19940815
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LI, LU, MC, NL, PT, SE
JP 09501693 T2 19970218 JP 1994-507626 19940815
US 5808006 A 19980915 US 1994-318628 19941011
US 5756672 A 19980526 US 1995-470108 19950606
PRIORITY APPLN. INFO.: US 1993-110664 19930820
WO 1994-US9120 19940815
AB A method for solubilizing and refolding peptides manufd. in bacterial hosts and accumulated as inclusion bodies is described. The method is optimized for the recovery of insulin-like growth factor and its analogs and is inexpensive. The polypeptide is resuspended at 0.1 - 15 mg/mL in a
buffer having a pH of about 7-12 of 5-40 vol.% of an alc. or polar aprotic solvent, about 0.2 to 3 M of an alk. earth, alkali metal, or ammonium salt, about 0.1 to 9 M of a ***chaotropic*** agent, and about 0.10 to 15 .mu.M of a copper or manganese salt. The protein is allowed to refold by incubating it in this buffer. The presence of the low concns. of copper or manganese minimizes the formation of incorrectly folded proteins
and avoids the need for disulfide exchange agents. The method can also be
used in two-phase systems where cell lysates are fractionated by phase partition and the phase contg. the inclusion bodies is under conditions suitable for solubilization and renaturation. The method is demonstrated with IGF-1 manufd. in Escherichia coli by expression of a cDNA. From a

large-scale fermn. (600-800 L) the protein could be refolded with a recovery of about 50%. Expts. using two-phase systems are reported.

=> d his

(FILE 'HOME' ENTERED AT 09:22:17 ON 24 MAR 2002)

FILE 'BIOSIS, EMBASE, MEDLINE, WPIDS, HCAPLUS' ENTERED AT 09:22:27 ON 24
MAR 2002
L1 327331 S PLASMID?
L2 72022 S BUTANOL?
L3 4823 S CHAOTROP?
L4 6 S L1 AND L2 AND L3
L5 4 DUP REM L4 (2 DUPLICATES REMOVED)

=> s l1 and l2
L6 201 L1 AND L2

=> s l6 not (cesium or cscl)
L7 200 L6 NOT (CESIUM OR CSCL)

=> s purif? or isolat? or separat?
4 FILES SEARCHED...
L8 5882048 PURIF? OR ISOLAT? OR SEPARAT?

=> s l7 and l8

L9 77 L7 AND L8

=> dup rem l9

PROCESSING COMPLETED FOR L9
L10 47 DUP REM L9 (30 DUPLICATES REMOVED)

=> d l10 ibib abs 1-47

L10 ANSWER 1 OF 47 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.DUPLICATE
1
ACCESSION NUMBER: 2002:130562 BIOSIS
DOCUMENT NUMBER: PREV200200130562
TITLE: Molecular characterization and transcriptional analysis of adhE2, the gene encoding the NADH-dependent aldehyde/alcohol dehydrogenase responsible for ***butanol*** production in alcohologenic cultures of Clostridium acetobutylicum ATCC 824.
AUTHOR(S): Fontaine, Lisa; Meynial-Salles, Isabelle; Girbal, Laurence; Yang, Xinghong; Croux, Christian; Soucaille, Philippe (1)
CORPORATE SOURCE: (1) Genencor International, 925 Page Mill Rd., Palo Alto, CA, 94304: psoucaille@genencor.com USA
SOURCE: Journal of Bacteriology, (February, 2002) Vol. 184, No. 3, pp. 821-830. http://intl-jb.asm.org/. print. ISSN: 0021-9193.
DOCUMENT TYPE: Article
LANGUAGE: English
AB The adhE2 gene of Clostridium acetobutylicum ATCC 824, coding for an aldehyde/alcohol dehydrogenase (AADH), was characterized from molecular and biochemical points of view. The 2,577-bp adhE2 codes for a 94.4-kDa protein. adhE2 is expressed, as a monocistronic operon, in alcohologenic cultures and not in solventogenic cultures. Primer extension analysis identified two transcriptional start sites 160 and 215 bp upstream of the adhE2 start codon. The expression of adhE2 from a ***plasmid*** in the DG1 mutant of C. acetobutylicum, a mutant cured of the pSOL1 megaplasmid, restored ***butanol*** production and provided elevated activities of NADH-dependent butyraldehyde and ***butanol*** dehydrogenases. The recombinant AdhE2 protein expressed in E. coli as a Strep-tag fusion protein and ***purified*** to homogeneity also demonstrated NADH-dependent butyraldehyde and ***butanol*** dehydrogenase activities. This is the second AADH identified in C. acetobutylicum ATCC 824, and to our knowledge this is the first example of a bacterium with two AADHs. It is noteworthy that the two corresponding genes, adhE and adhE2, are carried by the pSOL1 megaplasmid of C. acetobutylicum ATCC 824.

L10 ANSWER 2 OF 47 HCAPLUS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 2001:731007 HCAPLUS
DOCUMENT NUMBER: 135:271994
TITLE: Pseudomonas ipu operon and recombinant microorganisms for production of L-alaninol and .gamma.-glutamyl amides
INVENTOR(S): Leisinger, Thomas; van der Ploeg, Jan; Kiener, Andreas M.; Waesch, Susana Ivone de Azevedo; Maire, Tere
PATENT ASSIGNEE(S): Lonza A.-G., Switz.
SOURCE: PCT Int. Appl., 106 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: German
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2001073038	A2	20011004	WO 2001-EP3651	20010330

W: AE, AG, AL, AM, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG
PRIORITY APPLN. INFO.: EP 2000-106888 A 20000331
AB Disclosed are novel micro-organisms which are capable of transforming isopropylamine into L-alaninol and wherein the genes ipuH and ipuI coding for enzymes involved in the metabolization of L-alaninol are deactivated. The invention also relates to a method for the prodn. of L-alaninol or theanine using said novel micro-organisms. Thus, the ipuABCDEFGH operon of Pseudomonas was cloned and sequenced. A Pseudomonas ipuH-mutant was used to convert isopropylamine to L-alaninol. E. coli expressing the ipuABCDEFG genes also converted isopropylamine to L-alaninol. The ipuC gene was cloned and expressed in E. coli. The product, .gamma.-glutamylamide synthetase, was ***purified*** and shown to catalyze the formation of theanine from L-glutamic acid and ethylamine. A large no. of other amines were found to be suitable substrates.

L10 ANSWER 3 OF 47 HCAPLUS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 2001:636266 HCAPLUS
DOCUMENT NUMBER: 135:163349
TITLE: Rapid nucleic acid ***separation*** , ***isolation*** and ***purification*** methods
INVENTOR(S): Shao, Wen
PATENT ASSIGNEE(S): USA
SOURCE: PCT Int. Appl., 21 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2001062976	A1	20010830	WO 2001-US5575	20010222

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA,

CH, CN,
CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR,
HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU,
SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN,
YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY,
DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG
PRIORITY APPLN. INFO.: US 2000-184328P P 20000223
AB Provided are rapid and simple methods for ***isolation*** and ***purifn*** . of nucleic acid from crude aq. samples. The invention also provides a kit for ***isolation*** and ***purifn*** . of nucleic acids from an aq. sample.
REFERENCE COUNT: 3 THERE ARE 3 CITED REFERENCES AVAILABLE FOR THIS
RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L10 ANSWER 4 OF 47 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.DUPLICATE 2
ACCESSION NUMBER: 2001:567058 BIOSIS
DOCUMENT NUMBER: PREV200100567058
TITLE: Clostridium beijerinckii cells expressing Neocallimastix patriciarum glycoside hydrolases show enhanced lichenan utilization and solvent production.
AUTHOR(S): Lopez-Contreras, Ana M. (1); Smidt, Hauke; van der Oost, John; Claassen, Pieternel A. M.; Mooibroek, Hans; de Vos, Willem M.
CORPORATE SOURCE: (1) Laboratory of Microbiology, Hesselink van Suchtelenweg 4, 6703 CT, Wageningen: ana.lopez-contreras@algemeen.micr.wau.nl Netherlands
SOURCE: Applied and Environmental Microbiology, (November, 2001)
Vol. 67, No. 11, pp. 5127-5133. print.
ISSN: 0099-2240.
DOCUMENT TYPE: Article
LANGUAGE: English
SUMMARY LANGUAGE: English
AB Growth and the production of acetone, ***butanol*** , and ethanol by Clostridium beijerinckii NCIMB 8052 on several polysaccharides and sugars were analyzed. On crystalline cellulose, growth and solvent production were observed only when a mixture of fungal cellulases was added to the medium. On lichenan growth and solvent production occurred, but this polymer was only partially utilized. To increase utilization of these polymers and subsequent solvent production, the genes for two new glycoside hydrolases, celA and celD from the fungus Neocallimastix patriciarum, were cloned ***separately*** into C. beijerinckii. To do this, a secretion vector based on the pMTL500E shuttle vector and containing the promoter and signal sequence coding region of the Clostridium saccharobutylicum NCP262 eglA gene was constructed and fused either to the celA gene or the celD gene. Stable C. beijerinckii transformants were obtained with the resulting ***plasmids*** , pWUR3 (celA) and pWUR4 (celD). The recombinant strains showed clear halos on agar plates containing carboxymethyl cellulose upon staining with Congo red. In addition, their culture supernatants had significant endoglucanase activities (123 U/mg of protein for transformants harboring celA and 78 U/mg of protein for transformants harboring celD). Although C. beijerinckii harboring either celA or celD was not able to grow, ***separately*** or in mixed culture, on carboxymethyl cellulose or microcrystalline cellulose, both transformants showed a significant increase in solvent production during growth on lichenan and more extensive degradation of this polymer than that exhibited by the wild-type strain.

L10 ANSWER 5 OF 47 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.
ACCESSION NUMBER: 2001:452715 BIOSIS
DOCUMENT NUMBER: PREV200100452715
TITLE: Suppression of chemical mutagen-induced SOS response by alkylphenols from clove (Syzygium aromaticum) in the Salmonella typhimurium TA1535/pSK1002 umu test.
AUTHOR(S): Miyazawa, Mitsuo (1); Hisama, Masayoshi
CORPORATE SOURCE: (1) Department of Applied Chemistry, Faculty of Science and Engineering, Kinki University, Kowakae, Higashiosaka-shi, Osaka, 577-8502: miyazawa@apch.kindai.ac.jp Japan
SOURCE: Journal of Agricultural and Food Chemistry, (August, 2001)
Vol. 49, No. 8, pp. 4019-4025. print.
ISSN: 0021-8561.
DOCUMENT TYPE: Article
LANGUAGE: English
SUMMARY LANGUAGE: English
AB A methanol extract from clove (Syzygium aromaticum) showed a suppressive effect of the SOS-inducing activity on the mutagen 2-(2-furyl)-3-(5-nitro-2-furyl)acrylamide (furylfuramide) in the Salmonella typhimurium TA1535/pSK1002 umu test. The methanol extract was re-extracted with hexane, dichloromethane, ethyl acetate, ***butanol*** , and water. The hexane fraction showed a suppressive effect. Suppressive compounds in the hexane fraction were ***isolated*** by silica gel column chromatography and identified as trans-isoeugenol (1) and eugenol (2) by GC, GC-MS, IR, and 1H and 13C NMR spectroscopy. Compounds 1 and 2 suppressed the furylfuramide-induced SOS response in the umu test. Compounds 1 and 2 suppressed 42.3 and 29.9% of the SOS-inducing activity at a concentration of 0.60 mumol/mL. These compounds were assayed with other mutagens, 4-nitroquinolin 1-oxide (4NQO) and N-methyl-N'-nitro-N-nitrosoguanidine (MNNG). In addition, compounds 1 and 2 were assayed with aflatoxin B1 (AfB1) and 3-amino-1,4-dimethyl-5H-pyrido(4,3-b)indole (Trp-P-1), which require liver metabolizing enzymes. These compounds showed suppressive effects of the SOS-inducing activity against furylfuramide, 4NQO, AfB1, and Trp-P-1. To research the structure-activity relationship, methyl esters of 1 and 2 (1Me and 2Me) and o-eugenol (3), as compounds similar to 2, were also assayed with all mutagens. Compounds 1Me, 2Me, and 3 showed weak suppressive effects of the SOS-inducing activity against furylfuramide.

L10 ANSWER 6 OF 47 WPIDS COPYRIGHT 2002 DERWENT INFORMATION LTD DUPLICATE 3
ACCESSION NUMBER: 2000-387779 [33] WPIDS
DOC. NO. CPI: C2000-117783
TITLE: Extraction of ***plasmid*** DNA from DNA-containing material using an organic solvent, a chaotrope and water, useful particularly for obtaining ***plasmid*** DNA from bacterial culture.
DERWENT CLASS: B04 D16
INVENTOR(S): BUTT, N J; JONES, C P; JOENES, C P
PATENT ASSIGNEE(S): (CAMB-N) CAMBRIDGE MOLECULAR TECHNOLOGIES LTD; (WHAT-N) WHATMAN BIOSCIENCE LTD
COUNTRY COUNT: 89
PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
WO 2000029563	A1	20000525	(200033)*	EN	16

RW: AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW NL
OA PT SD SE SL SZ TZ UG ZW
W: AE AL AM AT AU AZ BA BB BG BR BY CA CH CN CU CZ DE DK EE ES FI GB
GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC LK LR LS LT LU
LV MA MD MG MK MN MW MX NO NZ PL PT RO RU SD SE SG SI SK SL TJ TM

TR TT TZ UA UG US UZ VN YU ZA ZW
GB 2346615 A 20000816 (200040)
AU 2000011699 A 20000605 (200042)
EP 1131420 A1 20010912 (200155) EN
R: AL AT BE CH CY DE DK ES FI FR GB GR IE IT LI LT LU LV MC MK NL PT
RO SE SI

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
WO 2000029563	A1	WO 1999-GB3830	19991117
GB 2346615	A	GB 1998-25215	19981117
AU 2000011699	A	AU 2000-11699	19991117
EP 1131420	A1	EP 1999-972256	19991117
		WO 1999-GB3830	19991117

FILING DETAILS:

PATENT NO	KIND	PATENT NO
AU 2000011699	A Based on	WO 200029563
EP 1131420	A1 Based on	WO 200029563

PRIORITY APPLN. INFO: GB 1998-25215 19981117
AN 2000-387779 [33] WPIDS
AB WO 200029563 A UPAB: 20010410
NOVELTY - A novel extraction mixture for selectively extracting ***plasmid*** DNA from a DNA -containing material, comprises a water-immiscible organic solvent capable of supporting ***plasmid*** DNA, a chaotrope and water.
DETAILED DESCRIPTION - A novel method for ***isolating*** ***plasmid*** DNA from a DNA containing material which comprises ***plasmid*** DNA and genomic DNA comprises:
(i) extracting the ***plasmid*** DNA into a water-immiscible organic solvent capable of supporting ***plasmid*** DNA, by mixing the material with the organic solvent, a chaotrope and water under conditions to denature the genomic DNA; and
(ii) recovering the ***plasmid*** DNA from the organic phase.
An INDEPENDENT CLAIM is also included for an extraction mixture for selectively extracting ***plasmid*** DNA from a water-immiscible organic solvent capable of supporting ***plasmid*** DNA, a chaotrope and water.
USE - The method can be used for the extraction of ***plasmid*** DNA from DNA-containing material such as a bacterial culture which may be lysed or unlysed.
ADVANTAGE - The method is capable of extracting ***plasmid*** DNA to high purity and with particularly low or zero contamination from genomic DNA.
Dwg.0/0

L10 ANSWER 7 OF 47 WPIDS COPYRIGHT 2002 DERWENT INFORMATION LTD
ACCESSION NUMBER: 2001-061875 [07] WPIDS
DOC. NO. CPI: C2001-017279
TITLE: ***Isolating*** extrachromosomal nucleic acids, by homogenizing sample, removing chromosomal nucleic acids, contacting sample with chaotropic agent to precipitate nucleic acids and recovering them.
DERWENT CLASS: A96 B04 D16
INVENTOR(S): DEDMAN, J R; KAETZEL, M A; REED, T D
PATENT ASSIGNEE(S): (UYCI-N) UNIV CINCINNATI
COUNTRY COUNT: 93
PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
WO 2000077235	A1	20001221	(200107)*	EN	44

RW: AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW MZ NL OA PT SD SE SL SZ TZ UG ZW
W: AE AG AL AM AT AU AZ BA BB BG BR BY CA CH CN CR CU CZ DE DK DM DZ EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC LK LR LS LT LU LV MA MD MG MK MN MW MX NO NZ PL PT RO RU SD SE SG SI SK SL TJ TM TR TT TZ UA UG US UZ VN YU ZA ZW
AU 2000056206 A 20010102 (200121)

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
WO 2000077235	A1	WO 2000-US16712	20000616
AU 2000056206	A	AU 2000-56206	20000616

FILING DETAILS:

PATENT NO	KIND	PATENT NO
AU 2000056206	A Based on	WO 200077235

PRIORITY APPLN. INFO: US 1999-139423P 19990616
AN 2001-061875 [07] WPIDS
AB WO 200077235 A UPAB: 20011129
NOVELTY - A new method for ***isolating*** extrachromosomal nucleic acids (NAs), comprises homogenizing a sample, removing chromosomal nucleic acids, contacting the sample with a chaotropic agent to precipitate nucleic acids and recovering nucleic acids
DETAILED DESCRIPTION - A novel method of ***isolating*** extrachromasomal NAs in a ***purified*** form comprises:
(a) homogenizing a biological sample;
(b) removing all of the chromosomal NAs from the biological sample;
(c) contacting the biological sample containing extrachromosomal NAs with a chaotropic solution under conditions which permit the NAs in the supernatants to precipitate; and
(d) recovering the extrachromosomal NAs.
An INDEPENDENT CLAIM is also included for a method of ***isolating*** NAs in a ***purified*** form comprising:
(i) homogenizing a biological sample containing NAs of interest;
(ii) partially ***purifying*** the NAs from the biological sample by adhering the NAs of interest to a retaining device;
(iii) contacting a gel containing the sample with a chaotropic solution under conditions which permit the NAs to precipitate; and
(iv) recovering the NAs.
USE - The method can be used for ***separating*** extracellular NAs from e.g. recombinant bacteriophage, ***plasmid*** , cosmid, yeast expression vector, viral expression vector, or retrovirus (claimed). The method can be used for ***isolating*** NAs in a cell system such as bacteria, yeast, insect, plant and mammalian systems (claimed).
ADVANTAGE - The methods can produce high yields of optimally pure intact extrachromosomal DNA without the need for toxic chemicals or DNA-binding materials. The process can be completed in 2-3 hours.
Dwg.0/0

L10 ANSWER 8 OF 47 WPIDS COPYRIGHT 2002 DERWENT INFORMATION LTD
ACCESSION NUMBER: 2000-505840 [45] WPIDS
DOC. NO. NON-CPI: N2000-374062
DOC. NO. CPI: C2000-151821
TITLE: Stable and efficient biological production of polyhydroxyalkanoates (PHA) containing 3-hydroxyhexanoate, comprising synthesis in transgenic organisms with transgene(s) encoding enzymes e.g. PHA polymerase.
DERWENT CLASS: A23 C06 D16 P13
INVENTOR(S): HUISMAN, G W; MADISON, L; PEOPLES, O P
PATENT ASSIGNEE(S): (META-N) METABOLIX INC
COUNTRY COUNT: 22
PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
WO 2000043523	A2	20000727	(200045)*	EN	48

RW: AT BE CH CY DE DK ES FI FR GB GR IE IT LU MC NL PT SE

W: AU CA JP MX
AU 2000026231 A 20000807 (200055)

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
WO 2000043523	A2	WO 2000-US1526	20000121
AU 2000026231	A	AU 2000-26231	20000121

FILING DETAILS:

PATENT NO	KIND	PATENT NO
AU 2000026231	A Based on	WO 200043523

PRIORITY APPLN. INFO: US 1999-235875 19990122
AN 2000-505840 [45] WPIDS
AB WO 200043523 A UPAB: 20000918
NOVELTY - Methods for production of polyhydroxyalkanoates (PHAs) containing 3-hydroxyhexanoate comprising synthesizing the PHA in transgenic organisms with at least 1 transgene encoding an enzyme, is new.

DETAILED DESCRIPTION - Methods for production of polyhydroxyalkanoates (PHAs) containing 3-hydroxyhexanoate comprising synthesizing the PHA in transgenic organisms with at least 1 transgene encoding an enzyme, is new. The enzyme is chosen from polyhydroxybutyrate (PHB) polymerase, PHA polymerase, beta -ketothiolase, beta -ketoacyl-coenzyme A (CoA) reductase, D-specific enoyl-CoA hydratase, crotonase, butyryl-CoA dehydrogenase and 3-hydroxybutyryl-CoA dehydrogenase integrated into the chromosome.

INDEPENDENT CLAIMS are also included for the following:
(1) methods of producing PHB-co-3-hydroxyhexanoate; and
(2) methods for producing 3-hydroxyhexanoate copolymers.

USE - The methods are used for the biological production of polyhydroxyalkanoates containing 3-hydroxyhexanoate (claimed), which are useful in molding applications particularly consumer packaging items such as bottles, cosmetic containers, diaper sheets, pens, golf tees and personal items such as molded tampon applicators. They provide genetically engineered systems for the production of polyhydroxyalkanoates containing 3-hydroxyhexanoates. They are used to provide useful mutations that can be used to produce 3-hydroxyhexanoic monomers from more economic feedstocks, such as butyrate or ***butanol*** . They are used to provide genes suitable for converting cellular metabolites derived from carbohydrate feedstocks to butyryl-CoA for the production of 3-hydroxyhexanoate co-monomers.

ADVANTAGE - The methods provide new pathways in biological systems for the endogenous synthesis of 3-hydroxyhexanoate co-monomers. They are genetically engineered biological systems for the production of PHAs containing 3-hydroxyhexanoate in which expression is sufficient and stable compared with prior-art ***plasmid*** -based systems.
Dwg.0/10

L10 ANSWER 9 OF 47 HCAPLUS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 2001:446380 HCAPLUS
DOCUMENT NUMBER: 135:16018
TITLE: Method for producing selenium-containing humanized abzyme
INVENTOR(S): Zhao, Daqing; Wang, Lin; Ding, Lan; Yan, Xiyun; Ni, Jiazuan
PATENT ASSIGNEE(S): Changchun Inst. of Applied Chemistry, Chinese Academy of Sciences, Peop. Rep. China
SOURCE: Faming Zhuanli Shenqing Gongkai Shuomingshu, 14 pp.
CODEN: CNXXEV
DOCUMENT TYPE: Patent
LANGUAGE: Chinese
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
CN 1274007	A	20001122	CN 1999-104234	19990513

OTHER SOURCE(S): MARPAT 135:16018
AB The invention provides a method for producing selenium-contg. humanized abzyme. The method comprises designing and synthesizing hapten; screening human single-chain antibody gene being specific to hapten; designing primer, amplifying, cloning to expression ***plasmid*** pET21a, expressing, sepg. and ***purifying*** to obtain single-chain antibody; and introducing catalytic group. The process for synthesis of hapten comprises substituting glutathione with 2,4-dinitrochlorobenzene at 10oC, esterifying with ROH in the presence of HCl at 4-20oC for 5-80 h, washing with Et ether, and drying. The process for screening single-chain antibody comprises coating immuno-plate with 10-40 .mu.g/mL hapten, adding human single-chain antibody phage library, standing at 15-37oC for 2 h, washing with 0.05M phosphate buffer (pH 7.2); adding 20-150 mM triethylamine, standing at 20-40oC for 8-15 min, neutralizing with 0.5-1.2 mM Tris-HCl buffer (pH 7.0-8.0), transfecting E.coli TG1 with OD of 0.2-1.0 at 20-40oC for 25-40 min, culturing in 100 .mu.g/mL ampicillin and 1% glucose-contg. culture medium 2XTY, and collecting antibody with feral phage M13K07. The process for introducing catalytic group selenium to antibody comprises allowing to react (0.4-1.2) x 10-5 mM antibody with 2.0-5.0 x 10-5 mM benzylsulfonyl fluoride in O2-free water at 15-30oC for 1-3 h, allowing to react with 0.05-0.25 mM NaHSe soln. at 30-40oC for 25-40 h under bubbling N2, and desalting on Sephadex G-10 column.

L10 ANSWER 10 OF 47 HCAPLUS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 2000:604091 HCAPLUS
DOCUMENT NUMBER: 134:323051
TITLE: Fast, non-toxic, and inexpensive n- ***butanol*** preparation of recombinant ***plasmids***
AUTHOR(S): Brieger, Jurgen; Weidt, Eberhard J.; Decker, Jochen
CORPORATE SOURCE: Department of Haematology and Oncology. III., Medical Division of the Johannes Gutenberg University, Mainz, 55131, Germany
SOURCE: Genetics and Molecular Biology (2000), 23(2), 299-300
CODEN: GMBIFG; ISSN: 1415-4757
PUBLISHER: Sociedade Brasileira de Genetica
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Various com. and non-com. ***plasmid*** prepn. protocols are currently available. However, the kits are expensive and many of the protocols contain toxic chems. Here we present a novel, optimized and, therefore, very advantageous ***plasmid*** prepn. protocol using n- ***butanol*** . The prepn. can be performed quickly and no toxic chems. are used, at overall costs of about one cent per ***plasmid*** prepn.
REFERENCE COUNT: 3 THERE ARE 3 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L10 ANSWER 11 OF 47 WPIDS COPYRIGHT 2002 DERWENT INFORMATION LTD DUPLICATE 4
ACCESSION NUMBER: 2000-194941 [17] WPIDS
DOC. NO. CPI: C2000-060362
TITLE: ***Isolating*** circular nucleic acids, particularly ***plasmids*** , from a mixture of nucleic acids comprises treating the mixture under alkaline conditions with a solid matrix in the presence of a chaotrophic substance.
DERWENT CLASS: B04 D16
INVENTOR(S): KANG, J; SAUER, P

PATENT ASSIGNEE(S): (QIAG-N) QIAGEN GMBH
COUNTRY COUNT: 27
PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
WO 9961603	A1	19991202 (200017)*		EN	32
RW: AT BE CH CY DE DK ES FI FR GB GR IE IT LU MC NL PT SE					
W: JP US					
EP 969090	A1	20000105 (200017)		EN	
R: AL AT BE CH CY DE DK ES FI FR GB GR IE IT LI LT LU LV MC MK NL PT					
RO SE SI					
EP 1088064	A1	20010404 (200120)		EN	
R: AT BE CH CY DE DK ES FI FR GB GR IE IT LI LU MC NL PT SE					

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
WO 9961603	A1	WO 1999-EP3660	19990527
EP 969090	A1	EP 1998-109593	19980527
EP 1088064	A1	EP 1999-938037	19990527
		WO 1999-EP3660	19990527

FILING DETAILS:

PATENT NO	KIND	PATENT NO
EP 1088064	A1 Based on	WO 9961603

PRIORITY APPLN. INFO: EP 1998-109593 19980527
AN 2000-194941 [17] WPIDS
AB WO 9961603 A UPAB: 20000405
NOVELTY - ***Separating*** and/or ***isolating*** circular nucleic acids from a mixture having different species of nucleic acids other than circular nucleic acids, comprises treating the mixture under alkaline conditions at a pH more than 8 with a solid matrix consisting essentially of a silica material in presence of at least one chaotropic substance.

DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for:

(1) an aqueous buffer comprising 6-9 M sodium thiocyanate, 0-20 vol.-% 1-4C-alcohols such as ethanol or isopropanol, 25-130 mM buffer substance preferably omega -amino acids; and

(2) a kit comprising the aqueous buffer of (1) and auxiliary materials such as columns with our without silicaceous material, suspensions of silicaceous material, additional buffers such as resuspension buffers, lysis buffers, washing buffers, elution buffers, instruction manual.

USE - For ***isolating*** and/or ***separating*** circular nucleic acids, particularly ***plasmids*** , from a mixture having different species of nucleic acids. ***Isolation*** of ***plasmid*** DNA is often a prerequisite for subsequent molecular biological experiments, e.g. PCR reactions, sequencing reactions, cloning reactions, restriction hydrolyses, transformations and transfections.

ADVANTAGE - Unlike all known methods for the ***isolation*** of ***plasmid*** DNA, the nucleic acids are ***isolated*** from bacterial crude lyzate avoiding the need to form cleared lyzate. (Clearing the lyzate is the most time-consuming step (e.g. by centrifugation) in ***plasmid*** ***isolation*** protocol or the major cost factor (e.g. by filtration)). The method is therefore simple and rapid.
Dwg.0/0

L10 ANSWER 12 OF 47 HCAPLUS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 1999:369353 HCAPLUS
DOCUMENT NUMBER: 131:166895
TITLE: Characterization of a Pseudomonas putida allylic alcohol dehydrogenase induced by growth on 2-methyl-3-buten-2-ol
AUTHOR(S): Malone, Vincent F.; Chastain, Amy J.; Ohlsson, John T.; Poneleit, Loelle S.; Nemecek-Marshall, Michele; Fall, Ray
CORPORATE SOURCE: Department of Chemistry and Biochemistry, and Cooperative Institute for Research in Environmental Sciences, University of Colorado, Boulder, CO, 80309-0215, USA
SOURCE: Appl. Environ. Microbiol. (1999), 65(6), 2622-2630
CODEN: AEMIDF; ISSN: 0099-2240
PUBLISHER: American Society for Microbiology
DOCUMENT TYPE: Journal
LANGUAGE: English
AB We have been working to develop an enzymic assay for the alc. 2-methyl-3-buten-2-ol (232-MB), which is produced and emitted by certain pines. To this end we have ***isolated*** the soil bacterium Pseudomonas putida MB-1, which uses 232-MB as a sole carbon source. Strain MB-1 contains inducible 3-methyl-2-buten-1-ol (321-MB) and 3-methyl-2-buten-1-al dehydrogenases, suggesting that 232-MB is metabolized by isomerization to 321-MB followed by oxidn. 321-MB dehydrogenase was ***purified*** to near-homogeneity and found to be a tetramer (151 kDa) with a subunit mass of 37,700 Da. It catalyzes NAD+-dependent, reversible oxidn. of 321-MB to 3-methyl-2-buten-1-al. The optimum pH for the oxidn. reaction was 10.0, while that for the redn. reaction was 5.4. 321-MB dehydrogenase oxidized a wide variety of aliph. and arom. alcs. but exhibited the highest catalytic specificity with allylic or benzylic substrates, including 321-MB, 3-chloro-2-buten-1-ol, and 3-aminobenzyl alc. The N-terminal sequence of the enzyme contained a region of 64% identity with the TOL ***plasmid*** -encoded benzyl alc. dehydrogenase of P. putida. The latter enzyme and the chromosomally encoded benzyl alc. dehydrogenase of Acinetobacter calcoaceticus were also found to catalyze 321-MB oxidn. These findings suggest that 321-MB dehydrogenase and other bacterial benzyl alc. dehydrogenases are broad-specificity allylic and benzylic alc. dehydrogenases that, in conjunction with a 232-MB isomerase, might be useful in an enzyme-linked assay for 232-MB.
REFERENCE COUNT: 36 THERE ARE 36 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L10 ANSWER 13 OF 47 HCAPLUS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 2000:751497 HCAPLUS
DOCUMENT NUMBER: 134:97621
TITLE: Degradation of organochlorine insecticide, endosulfan, in polluted soil through bacteria
AUTHOR(S): Shakoori, Farah R.; Rafique, Habib; Qureshi, Fatima; Ali, Syed Shahid; Shakoori, A. R.
CORPORATE SOURCE: Cell and Molecular Biology Laboratory, University of the Punjab, Lahore, Pak.
SOURCE: Proceedings of Pakistan Congress of Zoology (1999), 19, 283-298
CODEN: PKCZEK; ISSN: 1013-3461
PUBLISHER: Zoological Society of Pakistan
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Bacterial biodegrdn. of organochlorine insecticide, Endosulfan was studied in a selective medium devoid of glucose as carbon source, which was only provided in the form of insecticide. Four endosulfan degrading bacterial ***isolates*** , 2 of them designated CMBL I33 and I36 identified as Planococcus sp., 1 CMBL I34 identified as Marinococcus and 1 CMBL I35 identified as Acetobacter were ***isolated*** from the soil samples collected from Shahdara, Ichhra and agricultural fields at New Campus of the University of the Punjab, Lahore. Different growth conditions including the effect of pH and temp. were studied on their growth. The optimum pH of ***isolates*** CMBL I33 and I36 was found to be 8.5, while for CMBL I34 and 20 it was 7.0 and 7.5, resp. The optimum temp. for ***isolates*** CMBL I33 and 19 was 30.degree., while it was 37.degree. for ***isolates*** CMBL I35 and I36. Their growth curves showed typical pattern of growth. All the ***isolates*** showed growth on

heavy metals such as Pb2+, Cr6+, Cd2+. The bacterial strains ***isolated*** in the present study seem to be the strong candidates for their use in environmental bioremediation because of their ability to degrade organochlorine insecticide, endosulfan.

REFERENCE COUNT: 56 THERE ARE 56 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L10 ANSWER 14 OF 47 WPIDS COPYRIGHT 2002 DERWENT INFORMATION LTD
ACCESSION NUMBER: 1998-109819 [10] WPIDS
CROSS REFERENCE: 1992-066679 [09]; 1994-035067 [04]; 1994-101197 [12]; 1996-040226 [04]; 1997-447901 [41]; 1997-479456 [44]; 1997-558141 [51]; 1997-558142 [51]; 1998-158369 [14]; 1998-556391 [47]; 2000-593649 [54]
DOC. NO. CPI: C1998-036011
TITLE: New Pseudomonas gafA element - used to activate transcription of latent genes or those expressed at low level, useful for, e.g. inducing synthesis of antifungal agents.
DERWENT CLASS: B04 C05 C06 D16 E17
INVENTOR(S): GAFFNEY, T D; HILL, D S; LAM, S T; LIGON, J M; STEIN, J I
PATENT ASSIGNEE(S): (NOVS) NOVARTIS FINANCE CORP
COUNTRY COUNT: 1
PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
US 5710031	A	19980120 (199810)*			35

APPLICATION DETAILS:

PATENT NO	KIND		APPLICATION	DATE
US 5710031	A	CIP of	US 1990-570184	19900820
		CIP of	US 1992-908284	19920702
		CIP of	US 1993-87636	19930701
		CIP of	US 1994-258261	19940608
		Div ex	US 1994-287442	19940808
			US 1995-459174	19950602

FILING DETAILS:

PATENT NO	KIND		PATENT NO
US 5710031	A	CIP of	US 5639949
		Div ex	US 5670350

PRIORITY APPLN. INFO: US 1994-287442 19940808; US 1990-570184 19900820; US 1992-908284 19920702; US 1993-87636 19930701; US 1994-258261 19940608; US 1995-459174 19950602
AN 1998-109819 [10] WPIDS
CR 1992-066679 [09]; 1994-035067 [04]; 1994-101197 [12]; 1996-040226 [04]; 1997-447901 [41]; 1997-479456 [44]; 1997-558141 [51]; 1997-558142 [51]; 1998-158369 [14]; 1998-556391 [47]; 2000-593649 [54]
AB US 5710031 A UPAB: 20001109
New ***isolated*** gene-activating element (A) comprises a Pseudomonas gafA sequence (I) that induces expression of at least 1 gene (II) in a transformed Pseudomonas, where (II) is latent or expressed at low level. Also claimed are: (1) chimaeric expression constructs and ***plasmid*** vectors containing (A), and (2) transgenic Pseudomonas including (A).
USE - (A) may be used for introduction into Pseudomonas and activate expression of (II), useful for rendering Pseudomonas host effective against fungi pathogenic on plants (all claimed). The transformants express pyrrolnitrin, cyanide, chitinase and gelatinase which are active against Rhizoconia solani, Helminthosporium gramineae and some species of Pythium and Fusarium. (A) may also be used to ***isolate*** genes encoding anti-pathogenic substances (APS), active against, e.g. fungi, bacteria, nematodes and viruses, and very generally to transform cells other than Pseudomonas for increasing production of, e.g. antibiotics for pharmaceutical use, vitamins, growth factors, hormones, ***butanol*** , lactic acid, polysaccharides and enzymes. Transformed cells of Pseudomonas are applied at 50-5000 (especially 200-500) g/hectare.
ADVANTAGE - Transformed Pseudomonas have a broad spectrum of activity against plant pathogens and can compete more aggresively in the rhizosphere.
Dwg.0/1

L10 ANSWER 15 OF 47 HCAPLUS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 1998:568933 HCAPLUS
DOCUMENT NUMBER: 129:186145
TITLE: A haloalkane dehalogenase of Rhodococcus and the gene encoding it and use of the enzyme in treatment of waste halohydrocarbons
INVENTOR(S): Affholter, Joseph A.; Swanson, Paul E.; Kan, Hueylin L.; Richard, Ruth A.
PATENT ASSIGNEE(S): The Dow Chemical Company, USA
SOURCE: PCT Int. Appl., 89 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9836080	A1	19980820	WO 1998-US2776	19980213
W: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GE, GH, GM, GW, HU, ID, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, UZ, VN, YU, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, SD, SZ, UG, ZW, AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG				
AU 9863249	A1	19980908	AU 1998-63249	19980213
EP 970224	A1	20000112	EP 1998-907444	19980213
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI				
JP 2001512317	T2	20010821	JP 1998-535931	19980213

PRIORITY APPLN. INFO.: US 1997-38181P P 19970213
WO 1998-US2776 W 19980213
AB A haloalkane dehydrogenase of Rhodococcus ATCC 55388 that catalyzes the conversion of a halogenated aliph. substrate to the corresponding vicinal halohydrins is identified and characterized and the gene encoding it is cloned and expressed. The enzyme can be manufd. on a large scale for immobilization and use in the treatment of waste streams high in halogenated aliph. compds. The enzyme was ***purified*** chromatog. and amino acid sequence-derived primers used to amplify a primary clone. The gene was expressed in Escherichia coli using prior art expression vectors to manuf. the enzyme with N- or C-terminal affinity labels.

L10 ANSWER 16 OF 47 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.DUPLICATE 5
ACCESSION NUMBER: 1998:270378 BIOSIS
DOCUMENT NUMBER: PREV199800270378
TITLE: Recombinant flounder growth hormone from Escherichia coli: Overexpression, efficient recovery, and growth-promoting effect on juvenile flounder by oral administration.
AUTHOR(S): Jeh, Hoon Sung; Kim, Chun Hyung; Lee, Hong Kyun; Han, Kyuboem (I)

CORPORATE SOURCE: (1) Biotech Res. Inst., LG Chem Research Park, P.O. Box 61,
Yu Song, Science Town, Taejon 305-380 South Korea
SOURCE: Journal of Biotechnology, (Feb. 26, 1998) Vol. 60, No. 3, pp. 183-193.
ISSN: 0168-1656.
DOCUMENT TYPE: Article
LANGUAGE: English
AB An efficient production method for recombinant flounder growth hormone
(r-fGH) from Escherichia coli was developed and the biological activity of ***purified*** r-fGH was examined using juvenile flounder. The use of bicistronic construction in the expression ***plasmid*** resulted in the production of over 40% of the E. coli cellular protein as r-fGH. The r-fGH was recovered from cell lysates following inclusion body washing, solubilization and refolding in sodium dodecylsulfate (SDS) solution, and removal of contaminated proteins with secondary ***butanol*** treatment. The SDS content in ***purified*** r-fGH solution was adjusted to appropriate levels by diafiltration. More than 47% of the r-fGH was recovered from the E. coli cell lysates and the purity of recovered r-fGH was 98%. The oral administration of ***purified*** r-fGH to juvenile flounder, once a week for 4 weeks at a dosage of 40 mug
r-fGH g-1 fish body weight, resulted in significant increases both in weight and length. These results of overexpression, simple ***purification*** with high recovery yield and purity, and good growth-promoting activity of the r-fGH suggest that the production scheme
described in this study is useful for the potential application of r-fGH in fish farming.

L10 ANSWER 17 OF 47 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.DUPLICATE
6
ACCESSION NUMBER: 1998:92770 BIOSIS
DOCUMENT NUMBER: PREV199800092770
TITLE: Localization of nod, nif, and acidic exopolysaccharide determinants on a large ***plasmid*** in slow-growing cowpea Rhizobium sp. S2.
AUTHOR(S): Pandya, Snehal; Desai, Anjana (1)
CORPORATE SOURCE: (1) Biotechnol. Cent., Fac. Sci., M.S. Univ. Baroda, Baroda
390 002 India
SOURCE: Current Microbiology, (Jan., 1998) Vol. 36, No. 1, pp. 36-40.
ISSN: 0343-8651.
DOCUMENT TYPE: Article
LANGUAGE: English
AB Slow-growing cowpea Rhizobium sp. S2, a pigeon pea ***isolate*** , showed excessive synthesis of exopolysaccharide (EPS) and nod factor(s) only when grown in the presence of corresponding host root exudate (RE) or
naringenin (a flavonoid present in RE of piceon pea). muc- strain, a ***plasmid*** -cured derivative of the parent strain, showed negligible EPS production and failed to synthesize the nod factor(s) under similar growth conditions. The nod factor(s) was extracted and partially ***purified*** from the total EPS of the culture supernatant of S2 grown
in the presence of either RE or naringenin by the ***butanol*** -soluble, ethyl acetate-insoluble method. The parent strain also showed ex-planta nitrogenase activity, whereas in muc- it was not detectable. In contrast to observations of the location of nod, nif- and EPS-synthesizing genes on the chromosome in commonly studied cowpea Rhizobia and Bradyrhizobium sp. colony blot and dot blot hybridization studies revealed the presence of nod (common nod genes), nif (nif KDH); and EPS (pss) determinants to be on the large ***plasmid*** in the parent strain and therefore absent in muc-, which is a ***plasmid*** -cured derivative.

L10 ANSWER 18 OF 47 HCAPLUS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 1998:452193 HCAPLUS
DOCUMENT NUMBER: 129:184801
TITLE: Screening of recombinant DNA clones by single-colony micro-lysate/restriction enzyme analysis
AUTHOR(S): Peterson, Kenneth R.
CORPORATE SOURCE: Univ. Washington, Seattle, WA, USA
SOURCE: BioTechniques (1998), 25(1), 26, 28
CODEN: BTNQDO; ISSN: 0736-6205
PUBLISHER: Eaton Publishing Co.
DOCUMENT TYPE: Journal
LANGUAGE: English
AB A method is described for identifying correct transformants that couples DNA ***isolation*** from single colonies with restriction enzyme digestion. The protocol contains the following steps: (1) individual colonies are picked from transformation plates and resuspended in buffer; (2) ***plasmid*** DNA is released by ***butanol*** extn. and concd. by ethanol pptn.; (3) the DNA is then resuspended in a restriction endonuclease digestion mixt.; and (4) the restriction digests are fractionated by agarose gel electrophoresis.

L10 ANSWER 19 OF 47 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.DUPLICATE
7
ACCESSION NUMBER: 1998:135127 BIOSIS
DOCUMENT NUMBER: PREV199800135127
TITLE: Shuttle vectors for hyperthermophilic archaea.
AUTHOR(S): Aravalli, Rajagopal N.; Garrett, Roger A. (1)
CORPORATE SOURCE: (1) Inst. Molecular Biology, Univ. Copenhagen, Solvgade
83H, DK-1307 Copenhagen K Denmark
SOURCE: Extremophiles, (Nov., 1997) Vol. 1, No. 4, pp. 183-191.
ISSN: 1431-0651.
DOCUMENT TYPE: Article
LANGUAGE: English
AB Progress in understanding the basic molecular, biochemical, and physiological characteristics of archaeal hyperthermophiles has been limited by the lack of suitable expression vectors. Here, we report the construction of versatile shuttle vectors that can be maintained, and selected for, in both archaea and bacteria. The primary construct, pAG1, was produced by ligating portions of the archaeal cryptic ***plasmid*** pGT5 and the bacterial ***plasmid*** pUC19, both of which exhibit high
copy numbers. A second vector construct, pAG2, was generated, with a reduced copy number in Escherichia coli, by introducing the Rom/Rop gene
from pB R322 into pAG1. After transformation, both pAG1 and pAG2 were
stably maintained and propagated in the euryarchaeote Pyrococcus furiosus,
the crenarchaeote Sulfolobus acidocaldarius, and in Escherichia coli, An archaeal selective marker, the alcohol dehydrogenase gene from Sulfolobus
solfataricus, was ***isolated*** by polymerase chain reaction (PCR) amplification and cloned into the two constructs. They were stably maintained and expressed in the two archaea and conferred resistance to ***butanol*** and benzyl alcohol. However, the vector pAG21, deriving
from pAG2, proved the more stable in E. coli probably due to its lower copy number in the bacterium. Conditions are presented for the use of the vectors which, potentially, can be used for other hyperthermophilic archaea.

L10 ANSWER 20 OF 47 HCAPLUS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 1997:72368 HCAPLUS
DOCUMENT NUMBER: 126:88346
TITLE: Thioredoxin fusion proteins and their manufacture and the preparation and ***purification*** of interleukin 11 fusion products
INVENTOR(S): Vicik, Steven M.; Schauer, Neil L.; Mercer, James R.;
Levallie, Edward R.; Briasco, Catherine A.; Deetz, Jeffrey S.; Winters, Dwight; Thomas, Jenifer L.
PATENT ASSIGNEE(S): Genetics Institute, Inc., USA
SOURCE: PCT Int. Appl., 53 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9638570	A1	19961205	WO 1996-US4811	19960411

W: AU, CA, JP
RW: AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE
US 5760189 A 19980602 US 1995-464176 19950602

CA 2220447 AA 19961205 CA 1996-2220447 19960411
AU 9655378 A1 19961218 AU 1996-55378 19960411
AU 718013 B2 20000406
EP 828842 A1 19980318 EP 1996-912620 19960411
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT,
IE, FI
JP 11509725 T2 19990831 JP 1996-536447 19960411
PRIORITY APPLN. INFO.: US 1995-464176 19950602
WO 1996-US4811 19960411

AB Methods of using thioredoxin fusion proteins to simplify the release of proteins from a microbial host are described. The method is specifically intended for use in the manuf. of interleukin 11 and is suitable for large-scale processing. Efficient release of the protein is achieved by treatment of a cell suspension with a warm soln. of a chelating agent. Addn. of the soln. increases the temp. of the cell suspension by 20-40.degree.C (preferably 35.degree.C). An alc. soln. contg. a divalent cation is then added to solubilize the fusion protein. Preferably, EtOH is added to 14 vol% with the divalent cation a mixt. of Mg2+, Mn2+, and Ca2+ in the range 50-200 mM. Much of the unwanted material is pptd. by this soln. The fusion protein can then be pptd. with Zn2+ and further ***purified*** chromatog. The manuf. of an interleukin-11-thioredoxin fusion protein in Escherichia coli is demonstrated. The interleukin was released from the fusion protein by digestion with enterokinase to give an IL-11 protein with an endotoxin content of <0.1 ng/mg. The protein had the expected N-terminal sequence and peptide map. Optimization expts. for
release of the protein from Escherichia coli are reported.

L10 ANSWER 21 OF 47 MEDLINE
ACCESSION NUMBER: 97141149 MEDLINE
DOCUMENT NUMBER: 97141149 PubMed ID: 8987493
TITLE: The effect of novobiocin on solvent production by Clostridium acetobutylicum.
AUTHOR: Wong J; Bennett G N
CORPORATE SOURCE: Rice University, Department of Biochemistry and Cell
Biology, Houston, TX 77005-1892, USA.
SOURCE: JOURNAL OF INDUSTRIAL MICROBIOLOGY, (1996 Jun) 16 (6)
354-9.
Journal code: ALF; 8610887. ISSN: 0169-4146.
PUB. COUNTRY: ENGLAND: United Kingdom
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: B
ENTRY MONTH: 199702
ENTRY DATE: Entered STN: 19970227
Last Updated on STN: 19980206
Entered Medline: 19970213

AB Cells of Clostridium acetobutylicum treated with novoblocin, a DNA gyrase
inhibitor, produced higher butyrate levels and lower solvent levels with acetone being the most affected. Seven enzyme activities involved in acid and solvent production were analyzed. Among them, only CoA transferase,
required for acetone formation and acid uptake, experienced a significant decrease in activity. As in Escherichia coli and Bacillus subtilis, DNA from C. acetobutylicum became less negatively supercoiled in the early stationary phase (solventogenic stage), as shown by analysis of linking number of a reporter ***plasmid*** by agarose gel electrophoresis in the presence of chloroquine.

L10 ANSWER 22 OF 47 EMBASE COPYRIGHT 2002 ELSEVIER SCI. B.V.DUPLICATE 8
ACCESSION NUMBER: 96160852 EMBASE
DOCUMENT NUMBER: 1996160852
TITLE: Recombination-induced variants of Clostridium acetobutylicum ATCC 824 with increased solvent production.
AUTHOR: Wong J.; Bennett G.N.
CORPORATE SOURCE: Dept. of Biochemistry/Cell Biology, Rice University, 6100
Main Street,Houston, TX 77005-1892, United States
SOURCE: Current Microbiology, (1996) 32/6 (349-356).
ISSN: 0343-8651 CODEN: CUMIDD
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
SUMMARY LANGUAGE: English

AB Three sporulation-specific genes (orfA, sigE, sigG) from Clostridium acetobutylicum ATCC 824 are arranged in a cluster, encoding the putative .sigma.(E)-processing enzyme, .sigma.(E), and .sigma.(G) respectively. When they were transformed into Clostridium acetobutylicum while on a ***plasmid*** functional in this organism, transformants did not survive. Three kinds of recombinations were then attempted with nonreplicative ***plasmids*** : duplication of orfA and sigE, replacement of all of the three genes, and inactivation of orfA. While the wild-type strain ceased to grow and produce solvents in batch cultures after approximately 24 h, mutant strains were ***isolated*** that showed sustained growth for a much longer time and produced a threefold increase in acetone and ***butanol*** in test tube cultures. In addition, one of the derived strains showed a significantly higher growth rate. Features of the restriction maps of the recombinants did not correlate with expected maps, indicating possible complications occurring during the recombination events.

L10 ANSWER 23 OF 47 EMBASE COPYRIGHT 2002 ELSEVIER SCI. B.V.DUPLICATE 9
ACCESSION NUMBER: 96166076 EMBASE
DOCUMENT NUMBER: 1996166076
TITLE: General vectors for archaeal hyperthermophiles: Strategies based on a mobile intron and a ***plasmid*** .
AUTHOR: Aagaard C.; Leviev I.; Aravalli R.N.; Forterre P.; Prieur D.; Garrett R.A.
CORPORATE SOURCE: Institute Molecular Biology, Copenhagen University,
Solvgade 83H,DK-1307 Copenhagen, Denmark
SOURCE: FEMS Microbiology Reviews, (1996) 18/2-3 (93-104).
ISSN: 0168-6445 CODEN: FMREE4
COUNTRY: Netherlands
DOCUMENT TYPE: Journal; Conference Article
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
SUMMARY LANGUAGE: English

AB Although there are currently no cloning and expression vectors available for archaeal hyperthermophiles, small cryptic ***plasmids*** have been
characterized for these organisms as well as viruses and introns capable of spreading between cells. Below, we review the recent progress in adapting these genetic elements as vectors for Pyrococcus furiosus and Sulfolobus acidocaldarius. An efficient and reliable transformation procedure is described for both organisms. The potential of the mobile intron from Desulfurococcus mobilus, inserted into the bacterial vector pUC18 to generate a new type of vector, was investigated in S. acidocaldarius. A polylinker was inserted upstream from the open reading frame encoding the homing enzyme I-Dmol. Both the polylinker and a 276 bp
fragment of the tetracycline gene from pBR322 could be inserted into the intron- ***plasmid***. construct and spreading still occurred in the culture of S. acidocaldarius. Experiments are in progress to test the co-mobility of the alcohol dehydrogenase and .beta.-galactosidase genes from Sulfolobus species with the intron. A Shuttle vector pCSV1 was also produced by fusing the pGT5 ***plasmid*** from Pyrococcus abyssi and
the bacterial vector pUC19 which, on transformation, is stable in both organisms without selection. Growth inhibition studies indicate that both P. furiosus and S. acidocaldarius are sensitive to the antibiotics carbomycin, celesticetin, chloramphenicol and thiostrepton as well as ***butanol*** and butylic alcohol. Spontaneous mutants resistant to these drugs have been ***isolated*** carrying single site mutations in their 23S rRNA gene; they include mutants of S. acidocaldarius resistant to chloramphenicol, carbomycin and celesticetin with the mutation C2452U
and thiostrepton-resistant mutants of P. furiosus carrying the mutation A1067G (both numbers corresponding to Escherichia coil 23S rRNA). These
mutated genes are being developed as selective markers. Moreover, two .beta.-galactosidase genes from P. furiosus have been cloned as possible phenotypic markers; one of these exhibits maximum activity at 95.degree.C
with O-nitrophenyl .beta.-D-galactopyranoside as substrate.

L10 ANSWER 24 OF 47 MEDLINE
ACCESSION NUMBER: 95202076 MEDLINE
DOCUMENT NUMBER: 95202076 PubMed ID: 7894709

TITLE: Characterization and expression of the hydrogenase-encoding gene from Clostridium acetobutylicum P262.
AUTHOR: Santangelo J D; Durre P; Woods D R
CORPORATE SOURCE: Institut fur Mikrobiologie, Georg-August-Universitat, Gottingen, Germany.
SOURCE: MICROBIOLOGY, (1995 Jan) 141 (Pt 1) 171-80. Journal code: BXW; 9430468. ISSN: 1350-0872.
PUB. COUNTRY: ENGLAND: United Kingdom
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-U09760
ENTRY MONTH: 199504
ENTRY DATE: Entered STN: 19950504
Last Updated on STN: 19950504
Entered Medline: 19950427

AB The hydrogenase enzyme of Clostridium acetobutylicum plays a pivotal role in controlling electron flow, and hence carbon flow, during the complex biphasic fermentation of carbohydrates to the neutral solvents acetone and ***butanol*** . We report here the cloning and molecular characterization of the hydrogenase-encoding gene (hydA) from C. acetobutylicum P262. This gene was ***isolated*** by colony hybridization, using the Clostridium pasteurianum hydrogenase-1 gene as a probe. The DNA sequence encoding the hydA gene from C. acetobutylicum was determined, and revealed an ORF (1722 bp) encoding a 574 amino-acid protein. This C. acetobutylicum hydrogenase protein product has 82% similarity and 67% identity with the C. pasteurianum hydrogenase-1 protein. Northern blot analysis of RNA ***isolated*** from C. acetobutylicum indicates that the C. acetobutylicum hydrogenase protein product is translated from a monocistronic operon. RNA was ***isolated*** from the different morphological and physiological stages of a batch C. acetobutylicum fermentation, and further Northern blot analyses revealed no differences in the expression of the gene during acidogenesis as opposed to solventogenesis. Primer extension experiments confirmed these results and identified the 5' start of the mRNA transcript. These results correlated well with the physiological need for this organism to dispose of excess reducing equivalents.

L10 ANSWER 25 OF 47 HCAPLUS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 1994:527073 HCAPLUS
DOCUMENT NUMBER: 121:127073
TITLE: A highly sensitive method for detecting environmental insults by stress-induced bioluminescence in genetically engineered strains of Escherichia coli
INVENTOR(S): Larossa, Robert Alan; Majarian, William Robert; Van Dyk, Tina Kangas
PATENT ASSIGNEE(S): du Pont de Nemours, E. I., and Co., USA
SOURCE: PCT Int. Appl., 101 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9413831	A1	19940623	WO 1993-US11527	19931202
W: AU, CA, JP, KR, US, US				
RW: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE				
CA 2150232	AA	19940623	CA 1993-2150232	19931202
AU 9457304	A1	19940704	AU 1994-57304	19931202
EP 673439	A1	19950927	EP 1994-903321	19931202
EP 673439	B1	19970423		
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LI, LU, MC, NL, PT, SE				
JP 08504101	T2	19960507	JP 1993-514216	19931202
AT 152181	E	19970515	AT 1994-903321	19931202
ES 2102811	T3	19970801	ES 1994-903321	19931202
ZA 9309078	A	19950605	ZA 1993-9078	19931203
US 5683868	A	19971104	US 1994-244376	19941006

PRIORITY APPLN. INFO.: US 1992-988428 19921204
US 1993-63173 19930514
WO 1993-US11527 19931202

AB Subtle changes in environmental stress can be detected and measured at sublethal levels as a change in the bioluminescence output of a genetically engineered microorganism. The organism contains the bioluminescence lux gene complex under the control of a stress-inducible promoter. Transformation vectors for Escherichia coli RFM443 were constructed by direct cloning and PCR protocols included to amplify the stress-inducible promoter of interest. For example, the promoter of the stress gene grpE was ***isolated*** from ***plasmid*** pGrpE4 and fused to the promoterless lux gene operon ***isolated*** from ***plasmid*** pUCD615. Bioluminescent screening in response to stress induction by 1-8% ethanol indicated active grpE-lux fusion ligated DNA in the transformed E. coli. DNA contg. 11 other stress-inducible promoters, including the dnaK promoter from Lambda phage 9E4, was also amplified by PCR and used to construct transforming vectors. E. coli strains contg. a tolC- mutation, which alters the permeability of the cell envelope to hydrophobic chems., allowed a more sensitive bioluminescent response to sublethal concns. of org. environmental contaminants. A wide variety of org. and inorg. chem. compds. and environmental conditions (e.g., UV) induced bioluminescence from the various transformed E. coli strains.

L10 ANSWER 26 OF 47 HCAPLUS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 1994:451102 HCAPLUS
DOCUMENT NUMBER: 121:51102
TITLE: Cloning and high-level expression of the glutathione-independent formaldehyde dehydrogenase gene from Pseudomonas putida
AUTHOR(S): Ito, Kiyoshi; Takahashi, Mari; Yoshimoto, Tadashi; Tsuru, Daisuke
CORPORATE SOURCE: Sch. Pharmaceut. Sci., Nagasaki Univ., Nagasaki, 852, Japan
SOURCE: J. Bacteriol. (1994), 176(9), 2483-91
CODEN: JOBAAY; ISSN: 0021-9193
DOCUMENT TYPE: Journal
LANGUAGE: English

AB A DNA fragment of 485 bp was specifically amplified by PCR with primers based on the N-terminal sequence of the ***purified*** formaldehyde dehydrogenase (EC 1.2.1.46) from Pseudomonas putida and on that of a cyanogen bromide-derived peptide. With this product as a probe, a gene coding for formaldehyde dehydrogenase (fdhA) in P. putida chromosomal DNA was cloned in Escherichia coli DH5.alpha.. Sequencing anal. revealed that the fdhA gene contained a 1197-bp open reading frame, encoding a protein composed of 399 amino acid residues whose calcd. mol. wt. was 42,082. The transformant of E. coli DH5.alpha. harboring the hybrid ***plasmid*** , pFDHK3DN71, showed about 50-fold-higher formaldehyde dehydrogenase activity than P. putida. The predicted amino acid sequence contained several features characteristic of the zinc-contg. medium-chain alc. dehydrogenase (ADH) family. Most of the glycine residues strictly conserved within the family, including a Gly-Xaa-Gly-Xaa-Xaa-Gly pattern in the coenzyme binding domain, were well conserved in this enzyme. Regions around both the catalytic and the structural zinc atoms were also conserved. Analyses of structural and enzymic characteristics indicated that P. putida FDH belongs to the medium-chain ADH family, with mixed properties of mammalian class I and III ADHs.

L10 ANSWER 27 OF 47 EMBASE COPYRIGHT 2002 ELSEVIER SCI. B.V.
ACCESSION NUMBER: 93081939 EMBASE
DOCUMENT NUMBER: 1993081939
TITLE: High level functional expression of human .beta.1-adrenergic receptor in baculovirus-infected cells screened by a rapid in situ procedure.

AUTHOR: Ravet V.; Blin N.; Guillaume J.-L.; Petitjean F.; Cabanie L.; Strosberg A.D.
CORPORATE SOURCE: Lab Immuno-Pharmacologie Moleculaire, CNRS and Universite
Paris VII, Inst Cochin Genetique Moleculaire,Paris, France
SOURCE: Journal of Receptor Research, (1993) 13/1-4 (541-558).
ISSN: 0197-5110 CODEN: JRERDM
COUNTRY: United States
DOCUMENT TYPE: Journal; Conference Article
FILE SEGMENT: 029 Clinical Biochemistry
030 Pharmacology
037 Drug Literature Index
LANGUAGE: English
SUMMARY LANGUAGE: English
AB A novel screening assay for the identification of baculovirus infected cells expressing membrane receptors was developed by using a replica transfer technique. Sf9 cells were cotransfected with wild type baculoviral DNA and the transfer vector pVL941-.beta.1 containing the coding region of the human .beta.1-adrenergic receptor gene. Infected cells embedded in agarose were incubated with [125I]-iodocyanopindolol and
transferred onto filters that were subsequently autoradiographed. This procedure resulted in the ***isolation*** of recombinant baculoviruses that expressed .beta.1-adrenergic receptors. Binding assays carried out with [125I]-ICYP indicated that more than 600,000 receptors were expressed
per cell, the highest level noted so far for this receptor in genetically engineered cells. Sf9 cells expressing the .beta.1-AR were analysed by ligand binding, competition experiments, adenylyl cyclase stimulation and photoaffinity labeling. These cells express a homogenous population of receptors and display the known pharmacological properties of .beta.1-AR in human tissues.

L10 ANSWER 28 OF 47 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.DUPLICATE
10
ACCESSION NUMBER: 1993:428635 BIOSIS
DOCUMENT NUMBER: PREV199396083260
TITLE: A simple method for recovering DNA from agarose gels using
glass fibre filters.
AUTHOR(S): Verma, Vijeshwar; Qazi, Gulam N.
CORPORATE SOURCE: Genetic Eng. Unit, Regional Res. Lab., Canai Road, Jammu
Tawi-180 001 India
SOURCE: Biotechnology Techniques, (1993) Vol. 7, No. 7, pp. 469-471.
ISSN: 0951-208X.
DOCUMENT TYPE: Article
LANGUAGE: English
AB A simple and reproducible procedure for the recovery of ***plasmid***
DNA is described. The method was standardized for the ***purification***
of ***plasmids*** from Gluconobacter oxydans ATCC9937. The protocol is
based on the use of glass microfiber filter paper for entrapment of DNA and its subsequent recovery by an elution buffer. The method precludes the
use of phenol and ***butanol*** for the removal of proteins and ethidium bromide respectively, therefore, making the procedure inexpensive
and gentle.

L10 ANSWER 29 OF 47 HCAPLUS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 1992:3208 HCAPLUS
DOCUMENT NUMBER: 116:3208
TITLE: ***Separation*** of nucleic acids from biological samples with alcohols and protein denaturants
INVENTOR(S): Kamata, Kazuya; Satsuka, Toshiaki
PATENT ASSIGNEE(S): Tosoh Corp., Japan
SOURCE: Jpn. Kokai Tokkyo Koho, 6 pp.
CODEN: JKXXAF
DOCUMENT TYPE: Patent
LANGUAGE: Japanese
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
JP 03101688	A2	19910426	JP 1989-237327	19890914

AB Nucleic acids can be sepd. from samples (or fermn. cultures) by treating the sample with a protein denaturant (e.g. guanidine salt, urea, etc.) and then with .gtoreq.1 alc. (selected from e.g. EtOH, PrOH, BuOH, pentanol and hexanol) for pptn. or removal. No harmful solvent is useful in the extn. Thus, ***plasmid*** pIBI176-transformed Escherichia coli culture medium was treated with guanidine-HCl and stirred with iso-PrOH.
After centrifugation, the pptn. was washed with 70% EtOH twice, dissolved
in pH 8.0 Tris buffer contg. 1 mM EDTA and again centrifuged. The resulting supernatant was treated with EtOH and centrifuged to give DNA.

L10 ANSWER 30 OF 47 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.DUPLICATE
11
ACCESSION NUMBER: 1992:71822 BIOSIS
DOCUMENT NUMBER: BA93:40277
TITLE: A RAPID PROCEDURE FOR DETECTING RECOMBINANT
PLASMIDS USING ***BUTANOL*** EXTRACTION.
AUTHOR(S): MAK Y M; SORNARAJAH R; HO K K
CORPORATE SOURCE: BOTANY DEP., NATL. UNIVERSITY SINGAPORE, LOWER KENT RIDGE
ROAD, SINGAPORE 0511, REPUBLIC OF SINGAPORE.
SOURCE: BIOTECHNIQUES, (1991) 11 (6), 723.
CODEN: BTNQDO. ISSN: 0736-6205.
FILE SEGMENT: BA; OLD
LANGUAGE: English
AB A convenient procedure using ***butanol*** to extract DNA from bacteria is described. This procedure is suitable for rapidly screening a large number of bacterial cultures for recombinant ***plasmid*** DNA without the need for further ***purification*** and expensive chemicals.

L10 ANSWER 31 OF 47 MEDLINE DUPLICATE 12
ACCESSION NUMBER: 92172390 MEDLINE
DOCUMENT NUMBER: 92172390 PubMed ID: 1665338
TITLE: ***Isolation*** of restriction fragments from large ***plasmids*** recovered from bacteria with multiple ***plasmids*** .
AUTHOR: Via L E; Falkinham J O 3rd
CORPORATE SOURCE: Department of Biology, Virginia Polytechnic Institute and
State University, Blacksburg 24061-0406.
SOURCE: BIOTECHNIQUES, (1991 Oct) 11 (4) 442, 444.
Journal code: AN3; 8306785. ISSN: 0736-6205.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199204
ENTRY DATE: Entered STN: 19920424
Last Updated on STN: 19920424
Entered Medline: 19920408
AB A rapid and simple method for ***isolation*** of restriction DNA fragments from large ***plasmids*** is described. The loss of large ***plasmids*** is avoided by restriction endonuclease cleavage in an agarose gel before DNA precipitation. ***Plasmids*** were ***separated*** in low-melting-point agarose by electrophoresis, the desired ***plasmid*** DNA band was cut from the gel and digested with
a restriction endonuclease in the agarose. Restriction fragments in agarose were recovered by a modified phenol-extraction, concentrated with
2- ***butanol*** and precipitated with ethanol. The procedure simplifies the task of cloning genes from large ***plasmids*** , resulting in high yields of restriction fragments from a desired ***plasmid*** in a short time.

L10 ANSWER 32 OF 47 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.
ACCESSION NUMBER: 1992:27074 BIOSIS
DOCUMENT NUMBER: BA93:16349
TITLE: ***ISOLATION*** OF RESTRICTION FRAGMENTS

FROM LARGE
PLASMIDS RECOVERED FROM BACTERIA WITH MULTIPLE
PLASMIDS .
AUTHOR(S): VIA L E; FALKINHAM J O III
CORPORATE SOURCE: DEP. BIOLOGY, VA. POLYTECHNIC INST. STATE UNIV.,
BLACKSBURG, VA. 24061-0406.
SOURCE: BIOTECHNIQUES, (1991) 11 (4), 442,444.
CODEN: BTNQDO. ISSN: 0736-6205.
FILE SEGMENT: BA; OLD
LANGUAGE: English
AB A rapid and simple method for ***isolation*** of restriction DNA fragments from large ***plasmids*** is described. The loss of large ***plasmids*** is avoided by restriction endonuclease cleavage in an agarose gel before DNA precipitation. ***Plasmids*** were ***separated*** in low-melting-point agarose by electrophoresis, the desired ***plasmid*** DNA band was cut from the gel and digested with
a restriction endonuclease in the agarose. Restriction fragments in agarose were recovered by a modified phenol-extraction, concentrated with
2- ***butanol*** and precipitated with ethanol. The procedure simplifies the task of cloning genes from large ***plasmids*** , resulting in high yields of restriction fragments from a desired ***plasmid*** in a short time.

L10 ANSWER 33 OF 47 EMBASE COPYRIGHT 2002 ELSEVIER SCI. B.V.DUPLICATE 13
ACCESSION NUMBER: 92266087 EMBASE
DOCUMENT NUMBER: 1992266087
TITLE: Cloning of an NADH-dependent ***butanol*** dehydrogenase gene from Clostridium acetobutylicum.
AUTHOR: Petersen D.J.; Welch R.W.; Walter K.A.; Mermelstein L.D.;
Papoutsakis E.T.; Rudolph F.B.; Bennett G.N.
CORPORATE SOURCE: Department of Chemical Engineering, Northwestern
University,Evanston, IL 60208, United States
SOURCE: Annals of the New York Academy of Sciences, (1991) 646/-
(94-98).
ISSN: 0077-8923 CODEN: ANYAA
COUNTRY: United States
DOCUMENT TYPE: Journal; Conference Article
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
SUMMARY LANGUAGE: English
AB The acetone- ***butanol*** fermentation of C. acetobutylicum is characterized by the unique shift from acid to solvent production. The mechanism of the solventogenic switch involves the induction of several enzymes, including NADH-dependent ***butanol*** dehydrogenase (BDH) at
the onset of solventogenesis. This enzyme is responsible for the final conversion of butyraldehyde to ***butanol*** , and is distinct from the NADPH-dependent alcohol dehydrogenase (ADH) also present in the organism.
To characterize the genetic control of this gene, we have cloned and expressed it in E. coli. A .lambda.EMBL3 phage library of C. acetobutylicum DNA was screened via plaque hybridization using a [32P]-radiolabeled, 32-fold degenerate, 62-mer oligonucleotide probe. The probe was designed by reverse translation of the NH2-terminal amino acid sequence of ***purified*** BDH II. Southern blot experiments indicate that the phage insert was of clostridial origin and had no homology with the previously cloned NADPH- dependent ADH. Subcloning of DNA from
purified positive plaques has localized the gene to a 3.5-kb EcoRI
fragment from which the enzyme is well expressed. The sequence of the 25
NH2-terminal amino acids for the cloned enzyme ***purified*** from E.
coli was determined and found to be identical to that for the clostridial NADH-dependent BDH II. Maxicell analysis of [35S]- radiolabeled
plasmid -encoded proteins identified a species encoded by the clostridial insert with the expected M(r) of 42 kD.

L10 ANSWER 34 OF 47 MEDLINE
ACCESSION NUMBER: 91058577 MEDLINE
DOCUMENT NUMBER: 91058577 PubMed ID: 2173920
TITLE: Expression of rabbit cytochrome P-450IIE2 in yeast and stabilization of the enzyme by 4-methylpyrazole.
AUTHOR: Pernecky S J; Porter T D; Coon M J
CORPORATE SOURCE: Department of Biological Chemistry, Medical School,
University of Michigan, Ann Arbor 48109.
CONTRACT NUMBER: AA-06221 (NIAAA)
SOURCE: BIOCHEMICAL AND BIOPHYSICAL RESEARCH COMMUNICATIONS, (1990
Nov 15) 172 (3) 1331-7.
Journal code: 9Y8; 0372516. ISSN: 0006-291X.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199012
ENTRY DATE: Entered STN: 19910222
Last Updated on STN: 19990129
Entered Medline: 19901228
AB A rabbit cytochrome P-450IIE2 full-length cDNA was cloned into a yeast
episomal ***plasmid*** (YEp13) between the copper-responsive yeast metallothionein gene promoter (CUP1) and the iso-1-cytochrome c gene terminator (CYC1), and the cytochrome P-450 was expressed in Saccharomyces
cerevisiae. The microsomal fraction prepared from copper-treated cells exhibited a ferrous carbonyl difference spectrum with an absorption maximum at 451 nm and contained approximately 0.07 nmol of P-450IIE2 per
mg of protein. The P-450IIE2 protein expressed in yeast microsomes was catalytically competent as judged by the NADPH-dependent deethylation of
N-nitrosodiethylamine and by the oxidation of ***butanol*** . Cholate solubilization and polyethylene glycol fractionation of yeast microsomal P-450IIE2 yielded a preparation with a markedly lower specific content than that of intact microsomes, but, when 4-methylpyrazole was included during solubilization, the holoenzyme was completely stabilized.

L10 ANSWER 35 OF 47 MEDLINE DUPLICATE 14
ACCESSION NUMBER: 91099700 MEDLINE
DOCUMENT NUMBER: 91099700 PubMed ID: 2125292
TITLE: An efficient method for ***isolation*** of ***plasmid*** DNA from methylotrophic bacteria.
AUTHOR: Brenner V; Holubova I; Hubacek J
CORPORATE SOURCE: Institute of Microbiology, Czechoslovak Academy of
Sciences, Prague.
SOURCE: FOLIA MICROBIOLOGICA, (1990) 35 (5) 454-5.
Journal code: F23; 0376757. ISSN: 0015-5632.
PUB. COUNTRY: Czechoslovakia
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199102
ENTRY DATE: Entered STN: 19910329
Last Updated on STN: 19980206
Entered Medline: 19910219
AB A method, suitable for the ***isolation*** of closed circular ***plasmid*** DNA from methylotrophic bacteria is described. Improvement
of cell lysis was achieved by ***butanol*** extraction of cells before application of the lytic agent. Using this method, cryptic ***plasmids*** of 7.8, 14, 36 and 200 kb were ***purified*** from soil- ***isolated*** methylotrophs.

L10 ANSWER 36 OF 47 EMBASE COPYRIGHT 2002 ELSEVIER SCI. B.V.DUPLICATE 15
ACCESSION NUMBER: 90261662 EMBASE
DOCUMENT NUMBER: 1990261662
TITLE: Methods for cloning key primary metabolic enzymes and ancillary proteins associated with the acetone-***butanol*** fermentation of Clostridium acetobutylicum.
AUTHOR: Cary J.W.; Petersen D.J.; Bennett G.N.; Papoutsakis E.T.
CORPORATE SOURCE: Department of Biochemistry, Rice University, Houston, TX
77251-1892, United States

SOURCE: Annals of the New York Academy of Sciences, (1990) 589/-
(67-81).
ISSN: 0077-8923 CODEN: ANYAA
COUNTRY: United States
DOCUMENT TYPE: Journal; Conference Article
FILE SEGMENT: 022 Human Genetics
029 Clinical Biochemistry
LANGUAGE: English
SUMMARY LANGUAGE: English
AB The unavailability of genetically defined mutants for complementation has intensified the problems inherent in cloning genes from C. acetobutylicum. The uniqueness of some of the pathways of this organism coupled with the relative inefficiency of transformation of clostridia and few characterized mutants in these pathways have made cloning these genes by traditional complementation methods impractical. Oligonucleotide hybridization techniques have been shown to circumvent many problems involved in detecting protein expression. The ease of hybridization screening of plaques allows phage libraries to be examined more readily than is generally the case with colony screening techniques. Recombinant lambda phages also contain more DNA per insert than most ***plasmid*** vectors can maintain, thus further decreasing the amount of screening necessary. Cosmid libraries, offering even greater length of individual inserts, can be screened in a similar manner, although such screening incorporates the limitations of colony screening techniques. It is true that the technique hinges on the ability to obtain an amino acid sequence from which an oligonucleotide can be designed. In the past, the ability to obtain sequences was limited because the quantity and number of ***purified*** proteins were limited or the proteins were amino-terminally blocked. However, recent technological advances in this area, such as high-resolution gel ***separation*** techniques coupled with microsequencing, have opened the door to proteins previously inaccessible. Deformylation methods have been developed to deblock amino-terminally formylated proteins, and successful internal amino acid sequence analysis by in situ protease digestion has also been reported using only picomolar quantities of proteins ***separated*** by one- or two-dimensional gel electrophoresis. Protein and DNA sequence data banks have been significantly upgraded in the past few years. A proposed oligonucleotide sequence can be evaluated to determine what other possible sequences have similar homology; moreover, protein similarity comparisons between related species might possibly supplant the need for protein ***isolation*** if regions of highly conserved amino acid sequences are found. To our knowledge, this represents the first reported use of oligonucleotide probe hybridization screening technology as a strategy for cloning solvent pathway genes of C. acetobutylicum. Despite the deleterious effects on hybridization inherent in the high A + T content of C. acetobutylicum gene specific-directed oligonucleotides, the technique has been shown to function with few modifications to previously recorded systems.

L10 ANSWER 37 OF 47 HCAPLUS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 1990:84197 HCAPLUS
DOCUMENT NUMBER: 112:84197
TITLE: Low pH pharmaceutical compositions of recombinant beta-interferon
INVENTOR(S): Hershenson, Susan I.; Thomson, Jody
PATENT ASSIGNEE(S): Cetus Corp., USA
SOURCE: PCT Int. Appl., 35 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 8905158	A1	19890615	WO 1988-US4307	19881202
W: AU, DK, FI, JP, NO				
RW: AT, BE, CH, DE, FR, GB, IT, LU, NL, SE				
US 5004605	A	19910402	US 1987-131375	19871210
AU 8929287	A1	19890705	AU 1989-29287	19881202
CA 1335255	A1	19950418	CA 1988-585206	19881207

PRIORITY APPLN. INFO.: US 1987-131375 19871210
WO 1988-US4307 19881202
AB A stable parenteral compn. having pH of 2-4 comprises a recombinant interferon-.beta. (IFN-.beta.) dissolved in an inert carrier contg. glycerol or polyethylene glycol (mol. wt. 190-1600) as a stabilizer/solubilizer. The strain of IFN-.beta.ser17-producing Escherichia coli carrying ***plasmid*** -pSY2501 was cultured; the cells were disrupted and extd. with 2- ***butanol*** ; the ext. was ***purified*** by centrifugation, precolumn chromatog., ultrafiltration, and column chromatog.; the final supernatant was then stabilized by adding either 25% glycerol or 25% PEG-300. The formulations remained sol. after 1 wk storage at 4.degree.. Cytopathic effect assay results showed the formulations maintained bioactivity at 4.degree. during 1 wk.

L10 ANSWER 38 OF 47 HCAPLUS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 1990:4051 HCAPLUS
DOCUMENT NUMBER: 112:4051
TITLE: ***Purification*** and manipulation of DNA by immobilization on an insoluble matrix
INVENTOR(S): Dupret, Daniel; Sauvageot, Martine; Harsany, Veronique
PATENT ASSIGNEE(S): Appligene, Fr.
SOURCE: PCT Int. Appl., 16 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: French
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 8902436	A1	19890323	WO 1988-FR461	19880916
W: JP, US				
RW: AT, BE, CH, DE, FR, GB, IT, LU, NL, SE				
FR 2620706	A1	19890324	FR 1987-13010	19870917
FR 2623505	A1	19890526	FR 1987-16338	19871125
FR 2623505	B1	19910426		
EP 333813	A1	19890927	EP 1988-908250	19880916
R: AT, BE, CH, DE, FR, GB, IT, LI, LU, NL, SE				
JP 02501353	T2	19900517	JP 1988-507861	19880916

PRIORITY APPLN. INFO.: FR 1987-13010 19870917
FR 1987-16338 19871125
WO 1988-FR461 19880916
AB DNA in aq. solns. is bound to an insol. inorg. matrix (glass, ceramic, or metal) under appropriate ionic conditions. The matrix-bound DNA is then further ***purified*** by solvent extn. for delipidation, deproteinization, and removal of RNA, e.g. in a low-pressure chromatog. system. The immobilized DNA can also be chem. modified under anhyd. conditions. ***Plasmid*** DNA was recovered from cleared alk. lysates of Escherichia coli by the addn. of 2 vols. 8M NaI and an aliquot of glass powder. The glass powder was collected by centrifugation and washed with aliquots of water-satd. PhOH, CHCl3, and 50% EtOH. The DNA was eluted from the glass with dil. buffer. The DNA was free of protein, RNA, and chromosomal DNA contaminants.

L10 ANSWER 39 OF 47 MEDLINE
ACCESSION NUMBER: 89378762 MEDLINE
DOCUMENT NUMBER: 89378762 PubMed ID: 2673928
TITLE: Molecular analysis and nucleotide sequence of the adh1 gene encoding an NADPH-dependent ***butanol*** dehydrogenase in the Gram-positive anaerobe Clostridium acetobutylicum.
AUTHOR: Youngleson J S; Jones W A; Jones D T; Woods D R
CORPORATE SOURCE: Department of Microbiology, University of Cape Town, South Africa.
SOURCE: GENE, (1989 May 30) 78 (2) 355-64.
Journal code: FOP; 7706761. ISSN: 0378-1119.
PUB. COUNTRY: Netherlands
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-M26941

ENTRY MONTH: 198910
ENTRY DATE: Entered STN: 19900309
Last Updated on STN: 19900309
Entered Medline: 19891018
AB The nucleotide sequence of a 2081-bp fragment of Clostridium acetobutylicum DNA containing the adh1 gene was determined. The ***butanol*** dehydrogenase gene is referred to as the adh1 gene since it was shown to have activity using ***butanol*** and ethanol as substrates. The adh1 gene consisted of 1164 bp and encoded an alcohol dehydrogenase (ADH) enzyme of 388 aa residues with an Mr of 43,274. The adh1 gene was ***separated*** from an upstream open reading frame by an intergenic region of 354 bp. No promoter consensus sequences were identified in the intergenic upstream region and the adh1 gene did not appear to be expressed off its own promoter in Escherichia coli. Three ***separate*** types of ADH have been recognized. The ADH1 from C. acetobutylicum exhibited 39% homology with the Fe-containing ADH2 from Zymomonas mobilis and 37% homology with the ADH4 from Saccharomyces cerevisiae, but showed little or no homology with the other characterised types of ADH.

L10 ANSWER 40 OF 47 MEDLINE
ACCESSION NUMBER: 89123011 MEDLINE
DOCUMENT NUMBER: 89123011 PubMed ID: 2644186
TITLE: Novel aerobic tetracycline resistance gene that chemically modifies tetracycline.
AUTHOR: Speer B S; Salyers A A
CORPORATE SOURCE: Department of Microbiology, University of Illinois, Urbana 61801.
CONTRACT NUMBER: AI 22383 (NIAID)
SOURCE: JOURNAL OF BACTERIOLOGY, (1989 Jan) 171 (1) 148-53.
Journal code: HH3; 2985120R. ISSN: 0021-9193.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198903
ENTRY DATE: Entered STN: 19900308
Last Updated on STN: 19970203
Entered Medline: 19890313
AB A tetracycline resistance gene that was found originally on the Bacteroides ***plasmid*** pBF4 confers resistance on Escherichia coli but only when cells are growing aerobically. When E. coli EM24 carrying this aerobic tetracycline resistance (*Tcr) gene is grown in medium containing tetracycline, the resulting spent medium is no longer toxic to tetracycline-sensitive (Tcs) E. coli EM24 (B.S. Speer and A.A. Salyers, J. Bacteriol. 170: 1423-1429, 1988). To determine whether the *Tcr gene product modified tetracycline, we characterized the material resulting from incubation of E. coli (*Tcr) with tetracycline. When [7-3H(N)]tetracycline was added to cultures of E. coli (*Tcr), at least 90% of the label was recovered in the extracellular fluid. Therefore, tetracycline was not being sequestered by the cells. The labeled material behaved similarly to tetracycline with respect to solubility in various organic solvents. However, the UV-visible light spectrum had a single peak at 258 nm, whereas the tetracycline spectrum had a peak at 364 nm. The labeled material also had a faster migration rate than did tetracycline on thin-layer plates in a solvent system of ***butanol*** -methanol-10% citric acid (4:1:2, vol/vol/vol) and was separable from tetracycline by reverse-phase high-pressure liquid chromatography, using an acetronitrile-0.1% trifluoroacetic acid solvent system. These results demonstrate that the *Tcr gene product chemically modifies tetracycline. The *Tcr gene is the first example of a chemically modifying tetracycline resistance mechanism.

L10 ANSWER 41 OF 47 HCAPLUS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 1991:528542 HCAPLUS
DOCUMENT NUMBER: 115:128542
TITLE: Genetic manipulation of the clostridia
AUTHOR(S): Blaschek, Hans P.
CORPORATE SOURCE: Dep. Food Sci., Univ. Illinois, Urbana, IL, 61801, USA
SOURCE: Dev. Ind. Microbiol. (1989), 30, 35-42
CODEN: DIMCAL; ISSN: 0070-4563
DOCUMENT TYPE: Journal; General Review
LANGUAGE: English
AB A review with 59 refs. An overview of recent advances in the genetic manipulation of the clostridia is presented with specific ref. to the food poisoning microorganism, C. perfringens, and the acetone- ***butanol*** -ethanol producer, C. acetobutylicum. This involves an examn. into mutants and mutagenesis techniques, ***plasmid*** ***isolation*** techniques and ***plasmid*** -encoded functions, natural conjugation and transformation-based systems, shuttle vector development, as well as the cloning and expression of Clostridium genes. Specific ref. is made to the mobilization of a non-conjugative ***plasmid*** in C. perfringens, the recent development of an efficient (104 transformants/.mu.g DNA) electroporation-induced intact-cell transformation system for C. perfringens, the construction of an 8.0 kb Escherichia coli-C. perfringens shuttle vector (designated pAK201), and the further development and characterization of C. acetobutylicum alc.-tolerant and degenerant strains as well as glucose derepressed and hyperproducing amylolytic enzyme mutants. Preliminary studies on the cloning in E. coli HB101 of C. acetobutylicum SA-3 chromosomal DNA encoding starch hydrolytic activity is also discussed.

L10 ANSWER 42 OF 47 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.DUPLICATE 16
ACCESSION NUMBER: 1989:443861 BIOSIS
DOCUMENT NUMBER: BA88:92133
TITLE: RAPID ***ISOLATION*** OF HIGH-MOLECULAR-WEIGHT DNA FROM AGAROSE GELS.
AUTHOR(S): POLLMAN M J; ZUCCARELLI A J
CORPORATE SOURCE: DEP. MICROBIOL., LOMA LINDA UNIV. SCH. MED., LOMA LINDA, CALIF. 92350.
SOURCE: ANAL BIOCHEM, (1989) 181 (1), 12-17.
CODEN: ANBCA2. ISSN: 0003-2697.
FILE SEGMENT: BA; OLD
LANGUAGE: English
AB We have developed a simple, reliable, and rapid method for recovering DNA from agarose gels. While many methods for DNA extraction have already been described, few provide quantitative recovery of large DNA molecules. These procedures generally require costly apparatus, extended elution times, or considerable handling of the sample after elution. Our method employs a novel electroelution chamber constructed from acrylic plastic. Gel slices containing DNA are placed in the chamber between platinum electrodes. Voltage is applied and a continuous flow of buffer sweeps the eluted DNA from the chamber into an external receptacle. Elution is complete in 7 min. Concentrated DNA is obtained by ***butanol*** extraction and alcohol precipitation in 1 h. Recoveries, quantitated by counting radiolabeled DNA or by densitometry of analytical gels, were 94 to 100% for fragments of 4 to 50 kb. The eluted DNA was undergraded and could be digested with restriction enzymes, ligated, end-labeled, or used to transform cells as efficiently as noneluted DNA. Complete elution of a 100-kb ***plasmid*** , a 194-kb concatemer of bacteriophage .lambda., and of 440- and 550-kb chromosomes of Saccharomyces cerevisiae was also achieved using the same process. This method is suitable for routine use in a wide range of cloning applications, including the electrophoretic ***isolation*** of large DNA molecules.

L10 ANSWER 43 OF 47 HCAPLUS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 1989:179532 HCAPLUS
DOCUMENT NUMBER: 110:179532
TITLE: Pharmaceutical injections containing recombinant beta-interferons and nonionic surfactants and sugars as stabilizers
INVENTOR(S): Shaked, Zeev; Stewart, Tracy; Thomson, Jody; Thomson, James William; Taforo, Terrance; Hershenson, Susan
PATENT ASSIGNEE(S): Cetus Corp., USA
SOURCE: Eur. Pat. Appl., 83 pp.

CODEN: EPXXDW
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
EP 270799	A1	19880615	EP 1987-115693	19871026
EP 270799	B1	19940921		
R: AT, BE, CH, DE, ES, FR, GB, GR, IT, LI, LU, NL, SE				
CA 1294215	A1	19920114	CA 1987-549489	19871016
FI 8704706	A	19880428	FI 1987-4706	19871026
FI 95002	B	19950831		
FI 95002	C	19951211		
NO 8704445	A	19880428	NO 1987-4445	19871026
NO 175704	B	19940815		
NO 175704	C	19941123		
AU 8780191	A1	19880428	AU 1987-80191	19871027
DK 8705634	A	19880428	DK 1987-5634	19871027
JP 63179833	A2	19880723	JP 1987-271604	19871027
AU 9210633	A1	19920319	AU 1992-10633	19920203
AU 659127	B2	19950511		

PRIORITY APPLN. INFO.: US 1986-923423 19861027

AB A stable pharmaceutical compn. for parenteral administration comprises a recombinant interferon-.beta. (IFN-.beta.) protein dissolved in an inert carrier medium contg. .gtoreq.1 biocompatible nonionic polymeric detergent(s) as a solubilizer or stabilizer. The strain of IFN-.beta.ser17-producing E. coli carrying ***plasmid*** pSY2501 was cultured. The refractile bodies contg. IFN-.beta.ser17 protein were harvested, concd., and disrupted; EDTA was added to kill residual bacterial and sucrose was added to create a concn. gradient of 1.1-1.25 g/mL and the soln. was centrifuged. The pellet was solubilized in phosphate-buffered saline with 2% Na dodecyl sulfate and dithiothreitol and extd. with 2- ***butanol*** . The ext. was again mixed with 0.1% Na dodecyl sulfate in phosphate-buffered saline and dithiothreitol; the mixt. was centrifuged and the pellet contained 81% IFN-.beta.. The pellet was then processed by Sephacryl S200 pre-column chromatog., oxidized by o-iodobenzoic acid, concd., and ***purified*** by Sephadex G-75 column. The desalting step was performed at pH 9.2 with Sephadex G-25 column equilibrated with 0.1% Na laurate and the pH of the eluate was lowered to pH 3.0 to ppt. the Na laurate. The mixt. was centrifuged and the supernatant was stabilized by adding 0.15% Trycol LAL-12; the pH was raised to .apprx.7.0 with NaOH and 5g dextrose was then added; finally the soln. was sterile-filtered and the dosage amts. of IFN-.beta.ser17 (0.25 mg) were filled into vials and frozen to between -35 and -45.degree.. A preferred lyophilized normal dose formulation comprises recombinant IFN-.beta. 0.25 mg/mL, polyoxyethylene lauryl ether 0.15% by vol., Na phosphate buffer 20 mM, dextrose 0.2%, and mannitol 2%. These IFN-.beta. formulations are at least as stable as human serum albumin-contg. formulations and they do not require strong solubilizing agents such as SDS.

L10 ANSWER 44 OF 47 HCAPLUS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 1989:509835 HCAPLUS
DOCUMENT NUMBER: 111:109835
TITLE: Repeated sequences in chromosomal DNA of Streptomyces aureofaciens 2201
AUTHOR(S): Godany, A.; Pristas, P.; Muchova, J.; Zelinka, J.
CORPORATE SOURCE: Inst. Mol. Biol., Slovak Acad. Sci., Bratislava, 842 51, Czech.
SOURCE: Metab. Enzymol. Nucleic Acids Incl. Gene Manipulations, [Proc. Int. Symp.], 6th (1988), Meeting Date 1987, 167-71. Editor(s): Zelinka, Jan; Balan, Jozef. Plenum: New York, N. Y.
CODEN: 56OGAP
DOCUMENT TYPE: Conference
LANGUAGE: English

AB The repeated sequence of the chromosomal DNA of S. aureofaciens 2201 was found to consist of many copies of integrated ***plasmid*** pSA 2201 which encodes an antibiotic substance inhibitory toward Bacillus amyloliquefaciens and Micrococus luteus. Transformation of pSA 2201 DNA ***isolated*** from S. aureofaciens into Streptomyces lividans resulted in transformants which produced the antibiotic substance. A BamHI fragment of S. aureofaciens chromosomal DNA from the repeated sequence was cloned into pBR322 and ***plasmids*** contg. both orientations of the insert (pIMB 2001 and pIMB 2007) were used to transform S. lividans. Transformants with both orientations of the insert inhibited test microorganisms. ***Plasmid*** pIMB 2050 was also constructed by cloning the Tn5 kanamycin resistance gene and the larger XhoI fragment of repetitive chromosomal DNA of S. aureofaciens in pUC18. Escherichia coli and S. lividans transformed with this shuttle vector both produced the antibiotically active substance. The antibiotic substance was not released by E. coli; its prodn. was proved by ***butanol*** extn. ***Plasmid*** pIMB 2001, 2007, and 2050 DNAs were stable in E. coli, but were rather unstable in S. lividans 66 in which numerous deletions were obsd.

L10 ANSWER 45 OF 47 WPIDS COPYRIGHT 2002 DERWENT INFORMATION LTD
ACCESSION NUMBER: 1986-095679 [15] WPIDS
DOC. NO. CPI: C1986-040720
TITLE: Periplasmic mature protein e.g. HGH prodn. - using DNA encoding prokaryotic signals linked to DNA encoding mature eukaryotic proteins.
DERWENT CLASS: B04 D16
INVENTOR(S): BOCHNER, B R; CHANG, C; GRAY, G L; HEYNEKER, H L; MCFARLAND, N C; OLSON, K C; PAI, R; REY, M W; CHANG, C N; MCFARIAND, N C; PAI, R C
PATENT ASSIGNEE(S): (GETH) GENENTECH INC
COUNTRY COUNT: 13
PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
EP 177343	A	19860409 (198615)*	EN		62
R: AT BE CH DE FR GB IT LI LU NL SE					
JP 61092575	A	19860510 (198625)			
US 4680262	A	19870714 (198730)			
US 4963495	A	19901016 (199044)			
EP 177343	B1	19920722 (199230)	EN		39
R: AT BE CH DE FR GB IT LI LU NL SE					
DE 3586386	G	19920827 (199236)			
JP 06296491	A	19941025 (199502)			25
JP 08015440	B2	19960221 (199612)			28
JP 2521413	B2	19960807 (199636)			25

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
JP 61092575	A	JP 1985-222621	19851004
US 4680262	A	US 1984-658339	19841005
US 4963495	A	US 1984-658342	19841005
EP 177343	B1	EP 1985-307044	19851002
DE 3586386	G	DE 1985-3586386	19851002
		EP 1985-307044	19851002
JP 06296491	A Div ex	JP 1985-222621	19851004
		JP 1994-73169	19851004
JP 08015440	B2	JP 1985-222621	19851004
JP 2521413	B2 Div ex	JP 1985-222621	19851004
		JP 1994-73169	19851004

FILING DETAILS:

PATENT NO	KIND	PATENT NO
DE 3586386	G Based on	EP 177343
JP 08015440	B2 Based on	JP 61092575
JP 2521413	B2 Previous Publ.	JP 06296491

PRIORITY APPLN. INFO: US 1984-658342 19841005; US 1984-658095 19841005; US 1984-658339 19841005
AN 1986-095679 [15] WPIDS
AB EP 177343 A UPAB: 19970502
DNA encoding a prokaryotic secretion signal sequence, pref. an E. coli signal sequence other than that of beta-lactamase, such as an enterotoxin signal sequence, or an AP signal sequence, is operably linked at its 3' end to the 5' end of DNA encoding a mature enkaryotic protein other than chicken triose phosphate isomerase, pref. a mammalian protein such as HGH,
bovine growth hormone or porcine growth hormone.
A method for the periplasmic secretion of a mature eukaryotic protein in the periplasmic space of a host prokaryote comprises (a) constructing a vector for expressing a secretable direct hybrid, which vector contains DNA encoding a prokaryotic secretion signal sequence linked at its 3' end to the 5' end of DNA encoding the mature eukaryotic protein, (b) transforming a prokaryotic host with the vector, (c) culturing the transformed host and (d) allowing mature protein to collect in the periplasm of the host.
USE/ADVANTAGE - The vectors express hybrid preproteins in high yields
in host cells, cleave the signal sequence from the preprotein and secrete mature eukaryotic protein in the periplasmic space of the host cells. The HGH is used for the treatment of hypopituitary dwarfism, burns, wound healinq, dystrophy, bone knitting, diffuse gastric bleeding and pseudoarthrosis.
Dwg.0/6
ABEQ DE 3586386 G UPAB: 19930922
DNA encoding a prokaryotic secretion signal sequence, pref. an E. coli signal sequence other than that of beta-lactamase, such as an enterotoxin signal sequence, or an AP signal sequence, is operably linked at its 3' end to the 5' end of DNA encoding a mature enkaryotic protein other than chicken triose phosphate isomerase, pref. a mammalian protein such as HGH,
bovine growth hormone or porcine growth hormone.
A method for the periplasmic secretion of a mature eukaryotic protein in the periplasmic space of a host prokaryote comprises (a) constructing a vector for expressing a secretable direct hybrid, which vector contains DNA encoding a prokaryotic secretion signal sequence linked at its 3' end to the 5' end of DNA encoding the mature eukaryotic protein, (b) transforming a prokaryotic host with the vector, (c) culturing the transformed host and (d) allowing mature protein to collect in the periplasm of the host.
USE/ADVANTAGE - The vectors express hybrid preproteins in high yields
in host cells, cleave the signal sequence from the preprotein and secrete mature eukaryotic protein in the periplasmic space of the host cells. The HGH is used for the treatment of hypopituitary dwarfism, burns, wound healinq, dystrophy, bone knitting, diffuse gastric bleeding and pseudoarthrosis.
ABEQ EP 177343 B UPAB: 19930922
A hybrid DNA sequence encoding a protein having at least the amino terminal sequence of mature hGH operably linked to a DNA sequence encoding
an enterotoxin signal.
0/0
ABEQ US 4680262 A UPAB: 19930922
Protein (PR) is recovered from the periplasmic space of a bacterial cell transformed to secrete an eukaryotic PR by (A) contacting the cell with sufficient of a 2-4C alkanol, pref. EtOH or ***butanol*** , for a sufficient time to kill the cell without lysing the inner membrane, (B) freezing the cell and then thawing the cell and (C) recovering the periplasmic PR including the eukaryotic PR from the cell.
The cell is pref. heated to 35-55 deg.C for 0.5-20 mins., with the heating and contacting carried out simultaneously while the cell in an aq. suspension in the culture medium. The alkanol concn. is 0.5-10, esp. 1.5, vol.%. The suspension of thawed cell is diluted into a tris buffer. The PR is a mature eukaryotic PR. The cell is esp. E. coli and the PR is human growth hormone.
ADVANTAGE - Proteolytic degradation by proteases during recovery is
minimised as is contamination of the periplasmic PR by intracellular PR; a more viable, large scale process than known ones; use of contaminating proteinaceous reagents is avoided.
ABEQ US 4963495 A UPAB: 19930922
Recombinant DNA sequences that encode the formation of mature human growth
hormone are operably linked at the DNA region which encodes the terminal
amine gp. to a DNA sequence that encodes the STII signal.
Plasmids
for the transformation of suitable microorganisms have been ***isolated*** , e.g. pAP-STII-hGH, ptrp-STII-hGH and pAP-1.
USE - Escherichia coli are transformed and then selectively propagated to produce mature human growth hormone as a heterologous protein.

L10 ANSWER 46 OF 47 WPIDS COPYRIGHT 2002 DERWENT INFORMATION LTD
ACCESSION NUMBER: 1985-311996 [50] WPIDS
DOC. NO. CPI: C1985-134658
TITLE: ***Isolating*** and ***purifying*** alpha interferon - from genetically modified bacteria or animal cells, by partitioning between aq. urea and organic phase.
DERWENT CLASS: B04 D16
INVENTOR(S): LUDWIG, J; OBERMEIER, R; SALOMON, I
PATENT ASSIGNEE(S): (FARH) HOECHST AG
COUNTRY COUNT: 20
PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
EP 164069	A	19851211	(198550)*	GE	8
R: AT BE CH DE FR GB IT LI LU NL SE					
DE 3421302	A	19851212	(198551)		
AU 8543422	A	19851212	(198606)		
NO 8502313	A	19851230	(198608)		
JP 61009298	A	19860116	(198609)		
DK 8502544	A	19851209	(198610)		
FI 8502251	A	19851209	(198611)		
ZA 8504329	A	19851211	(198613)		
PT 80617	A	19860317	(198615)		
ES 8604022	A	19860601	(198631)		
US 4681931	A	19870721	(198731)		
EP 164069	B	19890329	(198913)	EN	
R: AT BE CH DE FR GB IT LI LU NL SE					
DE 3569123	G	19890503	(198919)		

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
EP 164069	A	EP 1985-106648	19850530
JP 61009298	A	JP 1985-122856	19850607
ZA 8504329	A	ZA 1985-4329	19850607
ES 8604022	A	ES 1985-543916	19850605
US 4681931	A	US 1985-741865	19850606

PRIORITY APPLN. INFO: DE 1984-3421302 19840608
AN 1985-311996 [50] WPIDS
AB EP 164069 A UPAB: 19930925
Plasmid -derived alpha-interferons (I) from genetically-modified bacterial cultures or (I) from culture supernatants of induced animal cells are ***isolated*** and ***purified*** by partitioning the crude material between (1) a surfactant-contg. aq urea soln and (2) a water-contg. n- ***butanol*** / acetic acid mixt. The ***purified*** (I) is then ***isolated*** from the upper phase.
USE/ADVANTAGE - The method is esp. used to recover (I) from genetically-engineered E.coli cells. It eliminates 95% of the impurities from the fermentation prod in a single stage and is simpler than known affinity or adsorption procedures.
0/0
ABEQ EP 164069 B UPAB: 19930925
A process for the ***isolation*** and ***purification*** of alpha-interferons generated by ***plasmids*** from bacterial cultures which have been modified by genetic engineering, and from culture supernatants from induced mammalian cells, which comprises partition of appropriate crude substances containing interferon between an aqueous urea
solution, which contains a surfactant, and a mixture of n- ***butanol*** and glacial acetic acid which contains water, and ***isolation*** of the ***purified*** alpha-interferons from the upper phase.
ABEQ US 4681931 A UPAB: 19930925

Isolation and ***purification*** of alpha-interferons comprises partition of crude materials between an aq. urea soln. contg. an anionic surfactant, and a mixt. of n- ***butanol*** and glacial acetic acid contg. water, followed by ***isolation*** of the ***purified*** prod. from the upper phase. The alpha-interferons are generated by ***plasmids*** from bacterial cultures which have been modified by genetic engineering, or are obtd. from culture supernatants of induced mammalian cells.

Pref. the lower phase contains 1-8 mol/l urea and 0.1-20% sodium dodecyl sulphate. Pref. upper phase is a mixt. of n- ***butanol*** / glacial acetic acid/water in ratio by vol. of 215:175:2150.

ADVANTAGE - Method enables elimination of more than 95% of all concomitant contaminants from E.coli fermentation, which is used for large scale industrial prodn. of alpha-interferons, for use as antivirals.

L10 ANSWER 47 OF 47 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.DUPLICATE 17

ACCESSION NUMBER: 1981:255875 BIOSIS
DOCUMENT NUMBER: BA72:40859
TITLE: ***PURIFICATION*** AND CHARACTERIZATION OF AN AUTO LYSIN FROM CLOSTRIDIUM-ACETOBUTYLICUM.
AUTHOR(S): WEBSTER J R; REID S J; JONES D T; WOODS D R
CORPORATE SOURCE: DEP. MICROBIOL., UNIV. CAPE TOWN, RONDEBOSCH 7700, S. AFR.
SOURCE: APPL ENVIRON MICROBIOL, (1981) 41 (2), 371-374. CODEN: AEMIDF. ISSN: 0099-2240.
FILE SEGMENT: BA; OLD
LANGUAGE: English

AB A proteinaceous substance with antibiotic-like activity, resembling that of a bacteriocin, was ***isolated*** from an industrial-scale acetone-***butanol*** fermentation of C. acetobutylicum. The substance, ***purified*** by acetone precipitation, DEAE chromatography and polyacrylamide gel electrophoresis, was characterized as a glycoprotein with a MW of 28,000. The glycoprotein was partially inactivated by certain protease enzymes. It had no effect on DNA, RNA or protein synthesis and it did not result in the loss of intracellular ATP. The glycoprotein lysed sodium dodecyl sulfate-treated cells and cell wall preparations, and therefore it is referred to as an autolysin. The autolysin gene appeared to be chromosomal since ***plasmid*** DNA was not detected in the C. acetobutylicum strain.

09/856336
AU #5

1. Document ID: US 20020001812 A1

L4: Entry 1 of 20

File: PGPB

Jan 3, 2002

PGPUB-DOCUMENT-NUMBER: 20020001812
PGPUB-FILING-TYPE: new
DOCUMENT-IDENTIFIER: US 20020001812 A1

TITLE: Mixed-bed solid phase and its use in the isolation of nucleic acids

PUBLICATION-DATE: January 3, 2002
US-CL-CURRENT: 435/6; 435/91.1, 436/17, 536/23.1

APPL-NO: 09/ 912045
DATE FILED: July 24, 2001

RELATED-US-APPL-DATA:
RLAN
RLFD
RLPC
RLKC
RLAC
09912045
Jul 24, 2001
GRANTED
A1
US
09312139
May 14, 1999
US
6270970

IN: Smith, Craig E., Holmes, Diana L., Simpson, Daniel J., Katzhendler, Jehoshua,
Bitner, Rex M., Grosch, Josephine C.

AB: Mixed-bed solid phases are provided, with methods for using such solid phases to
isolate target nucleic acids, such as plasmid DNA, chromosomal DNA, RNA, or nucleic acids
generated by enzymatic amplification from contaminants, including proteins, lipids, cellular
debris, or other nucleic acids. The mixed-bed solid phases of this invention are mixtures of at
least two different solid phases, each of which has a capacity to bind to the target nucleic
acid under different solution conditions, and the capacity to release the nucleic acid under
similar elution conditions. By exchanging solution conditions according to the methods of this
invention, one can remove contaminants from the target nucleic acid bound to the mixed-bed
solid phase, then elute the target nucleic acid in an elution buffer.

L4: Entry 1 of 20

File: PGPB

Jan 3, 2002

DOCUMENT-IDENTIFIER: US 20020001812 A1
TITLE: Mixed-bed solid phase and its use in the isolation of nucleic acids

BSTX:
[0009] Many conventional procedures for isolating target nucleic acids from various mixtures of the
target nucleic acids and contaminants, including mixtures produced from cells as described above,
entail the use of hazardous chemicals such as phenol, chloroform, and ethidium bromide. For example,
phenol or an organic solvent mixture containing phenol and chloroform are used in many such
conventional procedures to extract contaminants from mixtures of target nucleic acids and various
contaminants. Alternatively, cesium chloride-ethidium bromide gradients are used in place of or in
addition to phenol or phenol-chloroform extraction. Closed circular DNA, such as plasmid DNA,
intercalates with ethidium bromide and forms a band in a cesium chloride gradient formed after
several hours of ultracentrifugation. The DNA/ethidium bromide band is extracted therefrom and the
plasmid DNA isolated from the ethidium bromide using butanol or other conventional means. See, e.g.,
Molecular Cloning, ed. by Sambrook et al. (1989), pub. by Cold Spring Harbor Press, pp. 1.42-1.50.
The phenol/chloroform extraction step, or cesium chloride banding and ethidium bromide extraction
step is generally followed by precipitation of the nucleic acid material remaining in the extracted
aqueous phase by adding ethanol to that aqueous phase. The precipitate is typically removed from the
solution by centrifugation, and the resulting pellet of precipitate is allowed to dry before being
resuspended in water or a buffer solution for further processing or analysis.

2. Document ID: US 6270970 B1

L4: Entry 2 of 20

File: USPT

Aug 7, 2001

US-PAT-NO: 6270970
DOCUMENT-IDENTIFIER: US 6270970 B1
TITLE: Mixed-bed solid phase and its use in the isolation of nucleic acids
DATE-ISSUED: August 7, 2001

US-CL-CURRENT: 435/6; 435/803, 436/17, 530/334

APPL-NO: 9/ 312139
DATE FILED: May 14, 1999

IN: Smith; Craig E., Holmes; Diana L., Simpson; Daniel J., Katzenhendler; Jehoshua,
Bitner; Rex M., Grosch; Josephine C.

AB: Mixed-bed solid phases are provided, with methods for using such solid phases to
isolate target nucleic acids, such as plasmid DNA, chromosomal DNA, RNA, or nucleic acids
generated by enzymatic amplification from contaminants, including proteins, lipids, cellular
debris, or other nucleic acids. The mixed-bed solid phases of this invention are mixtures of at
least two different solid phases, each of which has a capacity to bind to the target nucleic
acid under different solution conditions, and the capacity to release the nucleic acid under
similar elution conditions. By exchanging solution conditions according to the methods of this
invention, one can remove contaminants from the target nucleic acid bound to the mixed-bed
solid phase, then elute the target nucleic acid in an elution buffer.

L4: Entry 2 of 20

File: USPT

Aug 7, 2001

DOCUMENT-IDENTIFIER: US 6270970 B1
TITLE: Mixed-bed solid phase and its use in the isolation of nucleic acids

BSPR:

Many conventional procedures for isolating target nucleic acids from various mixtures of the target nucleic acids and contaminants, including mixtures produced from cells as described above, entail the use of hazardous chemicals such as phenol, chloroform, and ethidium bromide. For example, phenol or an organic solvent mixture containing phenol and chloroform are used in many such conventional procedures to extract contaminants from mixtures of target nucleic acids and various contaminants.

Alternatively, cesium chloride-ethidium bromide gradients are used in place of or in addition to phenol or phenol-chloroform extraction. Closed circular DNA, such as plasmid DNA, intercalates with ethidium bromide and forms a band in a cesium chloride gradient formed after several hours of ultracentrifugation. The DNA/ethidium bromide band is extracted therefrom and the plasmid DNA isolated from the ethidium bromide using butanol or other conventional means. See, e.g., Molecular Cloning, ed. by Sambrook et al. (1989), pub. by Cold Spring Harbor Press, pp. 1.42-1.50. The phenol/chloroform extraction step, or cesium chloride banding and ethidium bromide extraction step is generally followed by precipitation of the nucleic acid material remaining in the extracted aqueous phase by adding ethanol to that aqueous phase. The precipitate is typically removed from the solution by centrifugation, and the resulting pellet of precipitate is allowed to dry before being resuspended in water or a buffer solution for further processing or analysis.

3. Document ID: US 6071713 A

L4: Entry 3 of 20

File: USPT

Jun 6, 2000

US-PAT-NO: 6071713
DOCUMENT-IDENTIFIER: US 6071713 A
TITLE: Bioprocesses for preparing 7-ACA and 7-ADAC
DATE-ISSUED: June 6, 2000

US-CL-CURRENT: 435/47; 435/183, 435/230, 435/243, 435/252.3, 435/254.11, 435/254.5, 435/49, 435/51, 435/935, 536/23.1, 536/23.2, 536/23.74

APPL-NO: 9/ 340781
DATE FILED: June 28, 1999

PARENT-CASE:

PRIOR APPLICATION This application is a division of U.S. patent application Ser. No. 839,327 filed Apr. 17, 1997, now U.S. Pat. No. 6,017,726 which is a division of U.S. patent application Ser. No. 439,404 filed May 11, 1995, now U.S. Pat. No. 5,629,171 which is a division of U.S. patent application Ser. No. 250,310 filed May 27, 1994, now U.S. Pat. No. 5,559,005 which is a continuation of U.S. patent application Ser. No. 07/953,492 filed Oct. 6, 1992, now abandoned which is a continuation-in-part of U.S. patent application Ser. No. 07/777,833 filed Oct. 15, 1991, now abandoned.

IN: Conder; Michael J., Rambosek; John A., McAda; Phyllis C., Reeves; Christopher D.

AB: A bioprocess for preparing adipoyl-7-ACA comprising the steps: (a) transforming cells of a strain of Penicillium chrysogenum which produces isopenicillin N with an expression vector containing DNA encoding an enzyme, having expandase activity capable of accepting adipoyl 6-APA as a substrate, an enzyme having hydroxylase activity capable of accepting adipoyl-7-ADCA as a substrate and an enzyme having acetyl transferase activity capable of accepting adipoyl 7-ADAC as a substrate; (b) culturing the transformed cells from step a) in a suitable culture medium containing an adipate feedstock, wherein said cells produce adipoyl 6-APA; and (c) culturing the transformed cells producing adipoyl 6-APA of step b) under conditions suitable for expression of said DNA encoding enzyme, thereby producing the end product adipoyl-7-ACA.

L4: Entry 3 of 20

File: USPT

Jun 6, 2000

DOCUMENT-IDENTIFIER: US 6071713 A
TITLE: Bioprocesses for preparing 7-ACA and 7-ADAC

DEPR:

E. coli cultures containing the plasmid of interest were grown in 500 mL LB broth (20 g/l of LB Broth Base (Gibco, Paisley, Scotland), with 15 .mu.g/mL tetracycline on a rotary shaker at 220 rpm for 12-16 hours at 37.degree. C. The cells were pelleted by centrifugation at 4000 xg for ten minutes at 4.degree. C. The cell pellet was resuspended in 18 mL Glucose Buffer (50 mM glucose, 25 mM Tris pH8.0, 10 mM EDTA) and 2 mL of 40 mg/mL lysozyme (Sigma, St. Louis, Mo.) in glucose buffer was added, mixed, and the mixture was incubated at room temperature for 15 minutes. Forty mL of a freshly prepared solution of 0.2N NaOH, 1% SDS was added, and the mixture swirled gently andplaced on ice for ten minutes. Thirty mL of 5M potassium acetate pH 4.8 were then added, mixed well, and the resultant mixture was placed on ice for an additional ten minutes. The cellular debris were pelleted by centrifugation at 4000 xg for ten minutes at 4.degree. C. and the resulting supernatant was filtered through a cheesecloth filter. Isopropanol (0.6 volumes) was added to the clarified supernatant to precipitate the plasmid DNA, and the precipitate was formed during incubation at room temperature for 20 minutes. The plasmid DNA was pelleted at 4000 xg for 20 minutes at 4.degree. C. and then washed with 70% ethanol and dried briefly. The pellet was resuspended in 9 mL TE buffer, then 10 grams of CsCl and 0.387 mL of a 10 mg/mL ethidium bromide solution were added. This solution was centrifuged at 313,100 xg for 24 hours. The resulting plasmid band in the cesium chloride gradient was visualized with ultraviolet light, isolated, and then the ethidium bromide was removed using water saturated butanol for extraction. The CsCl in the plasmid preparation was then removed by dialysis against TE buffer, and finally the DNA was concentrated using PEG (MW 8000).

Concentration of DNA was determined spectrophotometrically using an absorbance reading at 260 nm.

4. Document ID: US 6030638 A

L4: Entry 4 of 20

File: USPT

Feb 29, 2000

US-PAT-NO: 6030638
DOCUMENT-IDENTIFIER: US 6030638 A
TITLE: Plasmid for in vivo expression of prostaglandin synthase
DATE-ISSUED: February 29, 2000

US-CL-CURRENT: 424/450; 435/320.1, 435/325, 435/362, 435/375, 435/455

APPL-NO: 8/ 459493
DATE FILED: June 2, 1995

PARENT-CASE:
This application is a continuation of application Ser. No. 08/080,221, filed Jun. 21, 1993
abandoned, which is a continuation of application Ser. No. 07/746,941, filed Aug. 19, 1991
abandoned.

IN: Brigham; Kenneth, Conary; Jon T., Canonico; Angelo, Meyrick; Barbara

AB: A novel plasmid has inserted therein the cDNA for prostaglandin synthase. The
present invention further provides a method of increasing prostanoid production in vivo
including the general steps of delivering a prostaglandin synthase gene to cells in vivo and
hyperexpressing the gene to enhance prostanoid production in the cells.

L4: Entry 4 of 20

File: USPT

Feb 29, 2000

DOCUMENT-IDENTIFIER: US 6030638 A
TITLE: Plasmid for in vivo expression of prostaglandin synthase

DEPR:
Once a colony has been isolated which contains the piece of cloned DNA, a large scale plasmid
preparation is grown. The plasmid is purified by lysis of the bacteria and precipitation of the
plasmid with polyethylene glycol and then purified an additional time by ultracentrifugation in an
isopynic CsCl solution. After ultracentrifugation for 40 hours at 45,000 rpm, the purified plasmid
is withdrawn through the side of the tube and the ethidium bromide is removed by extraction with TE
saturated butanol. Finally, the isolated plasmid is precipitated with ethanol and resuspended in
sterile water. This double purification procedure (polyethylene glycol and CsCl) appears to be
critical to minimize endotoxin contamination.

5. Document ID: US 6017726 A

L4: Entry 5 of 20

File: USPT

Jan 25, 2000

US-PAT-NO: 6017726

DOCUMENT-IDENTIFIER: US 6017726 A
TITLE: Bioprocesses for preparing 7-ACA and 7-ADAC
DATE-ISSUED: January 25, 2000

US-CL-CURRENT: 435/47; 435/183, 435/2, 435/230, 435/320.1, 435/51, 435/935

APPL-NO: 8/ 839327
DATE FILED: April 17, 1997

PARENT-CASE:
PRIOR APPLICATIONS This application is a division of U.S. patent application Ser. No. 08/439,404
filed May 11, 1995, now U.S. Pat. No. 5,629,171 which is a division of U.S. patent application Ser.
No. 08/250,310 filed May 27, 1994, now U.S. Pat. No. 5,559,005 which is a continuation of U.S.
patent application Ser. No. 07/953,492 filed Oct. 6, 1992, now abandoned which is a
continuation-in-part of U.S. patent application Ser. No. 07/777,833 filed Oct. 15, 1991, now
abandoned.

IN: Conder; Michael J., Rambosek; John A., McAda; Phyllis C., Reeves; Christopher D.

AB: A process for making 7-aminodeacetyl-cephalosporanic acid (7-ADAC).

L4: Entry 5 of 20

File: USPT

Jan 25, 2000

DOCUMENT-IDENTIFIER: US 6017726 A
TITLE: Bioprocesses for preparing 7-ACA and 7-ADAC

DEPR:
E. coli cultures containing the plasmid of interest were grown in 500 mL LB broth (20 g/l of LB
Broth Base (Gibco, Paisley, Scotland), with 15 .mu.g/mL tetracycline on a rotary shaker at 220 rpm
for 12-16 hours at 37.degree. C. The cells were pelleted by centrifugation at 4000 xg for ten
minutes at 4.degree. C. The cell pellet was resuspended in 18 mL Glucose Buffer (50 mM glucose, 25
mM Tris pH 8.0, 10 mM EDTA) and 2 mL of 40 mg/mL lysozyme (Sigma, St. Louis, Mo.) in glucose buffer
was added, mixed, and the mixture was incubated at room temperature for 15 minutes. Forty mL of a
freshly prepared solution of 0.2N NaOH, 1% SDS was added, and the mixture swirled gently and placed
on ice for ten minutes. Thirty mL of 5M potassium acetate pH 4.8 were then added, mixed well, and
the resultant mixture was placed on ice for an additional ten minutes. The cellular debris were
pelleted by centrifugation at 4000 xg for ten minutes at 4.degree. C. and the resulting supernatant
was filtered through a cheesecloth filter. Isopropanol (0.6 volumes) was added to the clarified
supernatant to precipitate the plasmid DNA, and the precipitate was formed during incubation at room
temperature for 20 minutes. The plasmid DNA was pelleted at 4000 xg for 20 minutes at 4.degree. C.
and then washed with 70% ethanol and dried briefly. The pellet was resuspended in 9 mL TE buffer,
then 10 grams of CsCl and 0.387 mL of a 10 mg/mL ethidium bromide solution were added. This solution
was centrifuged at 313,100 xg for 24 hours. The resulting plasmid band in the cesium chloride
gradient was visualized with ultraviolet light, isolated, and then the ethidium bromide was removed
using water saturated butanol for extraction. The CsCl in the plasmid preparation was then removed
by dialysis against TE buffer, and finally the DNA was concentrated using

PEG (MW 8000).

Concentration of DNA was determined spectrophotometrically using an absorbance reading at 260 nm.

6. Document ID: US 5942427 A

L4: Entry 6 of 20

File: USPT

Aug 24, 1999

US-PAT-NO: 5942427
DOCUMENT-IDENTIFIER: US 5942427 A
TITLE: N-acetylmannosamine dehydrogenase gene and novel recombinant DNA as well as a method for
production of N-acetylmannosamine dehydrogenase
DATE-ISSUED: August 24, 1999

US-CL-CURRENT: 435/190; 435/252.33, 435/320.1, 435/69.1, 536/23.7

APPL-NO: 7/ 637865
DATE FILED: December 27, 1990

FOREIGN-APPL-PRIORITY-DATA:

COUNTRY	APPL-NO	APPL-DATE
JP	1-338267	December 28, 1989

IN: Otake; Hideko, Koyama; Yasuji, Horiuchi; Tatsuo, Nakano; Eiichi

AB: N-acetylmannosamine dehydrogenase gene derived from a microorganism belonging to the
genus Flavobacterium, e.g., Flavobacterium sp. No. 141-8 strain and defined by a specific
restriction enzyme map which encodes 271 amino acid sequence. Using the recombinant DNA,
N-acetylmannosamine dehydrogenase can be produced in a simpler manner in an industrial scale.
The enzyme is useful for quantitative determination of sialic acid.

L4: Entry 6 of 20

File: USPT

Aug 24, 1999

DOCUMENT-IDENTIFIER: US 5942427 A
TITLE: N-acetylmannosamine dehydrogenase gene and novel recombinant DNA as well as a method for
production of N-acetylmannosamine dehydrogenase

DEPR:
Then, the precipitates were subjected to a conventional drying treatment under reduced pressure,
which was then dissolved in 6 ml of 10 mM Tris-HCl buffer (pH 7.5) containing 1 mM EDTA. After 6 g
of cesium chloride and 0.2 ml of 10 mg/ml of ethidium bromide were added to the solution, the
mixture was subjected to equilibrium density gradient centrifugation at 39,000 rpm for 42 hours
using an ultra centrifugation machine to isolate recombinant plasmid pNAM 106 DNA. Ethidium bromide
was then removed with n-butanol, and dialysis was performed to 10 mM Tris-HCl buffer containing 1 mM
EDTA to give 800 .mu.g of purified recombinant plasmid pNAM 106 DNA.

7. Document ID: US 5847257 A

L4: Entry 7 of 20

File: USPT

Dec 8, 1998

US-PAT-NO: 5847257
DOCUMENT-IDENTIFIER: US 5847257 A
TITLE: Transgenic mouse deficient in T-cells
DATE-ISSUED: December 8, 1998

US-CL-CURRENT: 800/11; 435/320.1, 435/69.1, 435/69.4, 435/69.6, 435/91.2, 536/23.1, 536/23.5,
536/24.31, 800/18, 800/25

APPL-NO: 8/ 558651
DATE FILED: November 16, 1995

FOREIGN-APPL-PRIORITY-DATA:

COUNTRY	APPL-NO	APPL-DATE
KR	1994-30675	November 21, 1994

IN: Seo; Jeong-Sun, Kim; Soonhee, Park; Woong-Yang

AB: The present invention relates to a transgenic mouse deficient in T-cells, which is
provided by fusing human heat shock protein (Hsp) gene with H2K promoter and transferring it to
a mouse. Transgenic mouse line with a shrunken thymus and edficient in T-cells not having
mature T-cells can be obtained.

L4: Entry 7 of 20

File: USPT

Dec 8, 1998

DOCUMENT-IDENTIFIER: US 5847257 A
TITLE: Transgenic mouse deficient in T-cells

DEPL:
Step 1. Construction of Human Hsp Gene Expression Vector pH2.3 (Hunt, C. & Morimoto, R. I., 1985,
Proc, Natl. Acad. Sci. 82) was digested with BamHI and HindIII restriction enzyme to obtain human
Hsp gene, and thereby a 2.3 kb DAN fragment was obtained. pH2K (Morello, D. et al, 1986, EMBO. J. 5,
1877-1883) was treated with HindIII and EcoRI to obtain 2.0 kb of H2K promoter. Two fragments were
ligated to obtain 4.3 kb gene fragment (FIG. 1), then pH2K/HSP (FIG. 2) was prepared by cloning the
4.3 kb fragment on restriction enzyme (ECoRI and Hind III) site of pUC19 purchased from New England
Biolabs. After transforming HB 101 E.coli by inserting pH2K/HSP to HB 101 E. Coli and the
transformed E.coli was stored. The above E.coli was inoculated to ampicillin-containing LB medium
(1% Bactotrypton, 0.5% Yeast Extract, 1% NaCl) to prepare a DNA solution for microinjection and
cultured overnight at 37.degree. C. The precipitate was centrifuged for 10 minutes at 2000 g, the

resultant precipitate was collected, and dissolved in the solution of 50 mM glucose, 10 mM Tris-Cl,
1 mM EDTA, 4 mg/ml lysozyme and kept on the ice for 5 minutes. This solution was carefully mixed
with 2 volumes of 0.2N NaOH and 1% SDS solution, kept for 5 minutes at room temperature, mixed with
0.5 volume of 5M potassium acetate solution, and kept on the ice for 5 minutes. The supernatant was
obtained by centrifugation at 12000 g for 10 minutes. The supernatant was extracted with phenol and
phenol / chloroform / isoamylalcohol (25:24:1) two or three times, and plasmid DNA was precipitated
by adding 0.1 volume of 3M sodium acetate and 2 volumes of ethanol. The precipitated DNA was
dissolved in TE buffer (10 mM Tris-CL, pH 8.0/1 mM EDTA, pH 8.0) and CsCl was added to have 1 g/ml
of final concentration of CsCl. This solution was centrifuged at 100,000 rpm for 12 hours by Beckman
TL 100 rotor for purification of DNA. The band including plasmid DNA was separated with an injector,
extracted with n-butanol and dialysed for 24 hours against TE buffer. After plasmid DNA extraction,
DNA was digested with restriction enzyme EcoRI, purified by excising agarose gel fragment containing
H2K-Hsp 70 gene of 4.3 kb after electrophoresis on 0.8% agarose gel. The concentration of DNA was
quantified and DNA was diluted with 10 mM Tris-Cl, pH 7.5/0.2 mM EDTA, pH 8.0 buffer to become 4
ng/.mu.l of DNA, and the DNA was stored at -20.degree. C. to be used as an injector.

8. Document ID: US 5824840 A

L4: Entry 8 of 20

File: USPT

Oct 20, 1998

US-PAT-NO: 5824840
DOCUMENT-IDENTIFIER: US 5824840 A
TITLE: Diabetes-inducing transgenic mouse
DATE-ISSUED: October 20, 1998

US-CL-CURRENT: 800/9; 435/320.1, 435/69.1, 435/69.6, 435/69.7, 435/91.2, 536/23.1, 536/23.5,
536/24.31, 800/22, 800/25

APPL-NO: 8/ 558719
DATE FILED: November 16, 1995

FOREIGN-APPL-PRIORITY-DATA:

COUNTRY	APPL-NO	APPL-DATE
KR	94-30676	November 21, 1994

IN: Seo; Jeong-Sun, Kim; Soonhee, Kim; Jongil

AB: Disclosed in this invention is a transgenic mouse containing rcombinant DNA
including a promoter and a heat shock protein 70 gene attached to downstream of the promoter.
Transgenic mice line inducing non-insulin dependent diabetes having a blood glucose level of
300 mg/dl was obtained.

L4: Entry 8 of 20

File: USPT

Oct 20, 1998

DOCUMENT-IDENTIFIER: US 5824840 A
TITLE: Diabetes-inducing transgenic mouse

DEPR:
pSP65INS (N. Sarvetnick et al., 1988, Cell vol.52, 773-7882) was treated with EcoRI and BamHI
restriction enzyme to obtain 1.8 kb human insulin promoter. Two fragments were ligated to obtain 4.1
KB fragment FIG. 1) and the fragment was cloned on EcoRI and HindIII restriction enzyme site of
pSP65 purchased from Promega to prepare pINS/HSP FIG. 2), and then the pINS/HSP was introduced into
HB 101 E. coli to obtain transformed strains. In order to prepare DNA solution for microinjection,
the E. coli was inoculated with LB medium (1% Bactotrypton, 0.5% Yeast Extract, 1% NaCl) containing
ampicillin, cultured at a temperature of 37.degree. C. overnight. The culture was collected by
centrifuging the solution at 2000 g for 10 minutes, and dissolving in a solution containing 50 mM
glucose, 10 mM Tris-Cl, 1 mM EDTA, 4 mg/ml lysozyme, and allowing same to incubate for 5 minutes in
an ice bath. After 0.2N NAOH, 2 volume of 1% SDS solution were cautiously mixed with the resulting
solution, the mixture was shaken and allowed to settle at room temperature for 5 minutes.
Thereafter, 0.5 volumes of 5M potassium acetate solution was added to the solution, and the obtained
mixture was allowed in an ice bath, and centrifuged at 12,000 for 10 minutes to obtain supernatant.
The supernatant was extracted two or three times with phenol and phenol/chroloform/isoamylalcohol
(28:24:1), and plasmid DNA was precipitated by adding 0.1 volumes of 3M sodium acetate and 2 volumes
of ethanol to the extract. The precipitated DNA was dissolved in TE buffer solution (10 mM Tris-Cl,
pH 8.0/1 mM EDTA, pH 8.0), and CsCl was added to the solution to have 1 g/ml of final concentration
of CsCl. The solution was centrifiged at 100,000 rpm for 12 hours by Beckman TL 100 roter to obtain
purified plasmid DNA. The band with the plasmid DNA was separated using a syringe, extracted with
n-butanol, dialysed against TE buffer solution for 24 hours. After plasmid DNA extraction, DNA was
digested with Hind III restriction enzyme, and electrophoresed through 0.8% agarose gel, and DNA was
purified by cutting the agarose gel fragment containing 4.1 KB DNA inducing diabetes. The
concentration of the DNA was determined, and then the DNA was diluted with the TE buffer solution
containing 10 mM of tris-Cl, pH 7.5/0.2 mM EDTA, pH 8.0 to concentration of DNA of 4 ng/ul, and the
DNA stored at a temperature of -20.degree. C. and used for microinjection.

9. Document ID: US 5665584 A

L4: Entry 9 of 20

File: USPT

Sep 9, 1997

US-PAT-NO: 5665584
DOCUMENT-IDENTIFIER: US 5665584 A
TITLE: DNA fragment containing a tannase gene, a recombinant plasmid, a process for producing
tannase, and a promoter
DATE-ISSUED: September 9, 1997

US-CL-CURRENT: 435/197; 435/320.1, 536/23.2, 536/24.1

APPL-NO: 8/ 460860
DATE FILED: June 5, 1995

FOREIGN-APPL-PRIORITY-DATA:
COUNTRY
APPL-NO
APPL-DATE
JP
6-159973
July 12, 1994
JP
7-083973
April 10, 1995

IN: Hatamoto; Osamu, Watarai; Teruo, Mizusawa; Kiyoshi, Nakano; Eiichi

AB: The present invention relates to a DNA fragment of 3,563 base pairs containing a
gene coding for tannase and derived from a microorganism belonging to the genus Aspergillus,
with the following restriction enzyme map: ##STR1## B: Bam HI, H: Hind III, K: Kpn I, S: Sal I,
X: Xba I; a DNA fragment containing a tannase gene coding for the amino acid sequence of (SEQ
ID NO:4); a recombinant plasmid comprising the DNA fragment containing the tannase gene
inserted into a plasmid vector; a process for producing tannase, comprising culturing a
microorganism belonging to the genus Aspergillus capable of producing tannase in medium with
the recombinant plasmid, and recovering tannase from the culture; and a promoter represented by
the nucleotide sequence of (SEQ ID NO:1). Tannase can be efficiently produced according to the
present invention.

L4: Entry 9 of 20
File: USPT
Sep 9, 1997

DOCUMENT-IDENTIFIER: US 5665584 A
TITLE: DNA fragment containing a tannase gene, a recombinant plasmid, a process for producing
tannase, and a promoter

DEPR:
The precipitate was dried under reduced pressure and dissolved in 6 ml TE buffer, followed by
addition of 6 g cesium chloride and 0.3 ml of 10 mg/ml ethidium bromide. This sample was separated
by equilibrium density-gradient centrifugation at 50,000 r.p.m. for 20 hours in a ultracentrifuge.
From the recombinant plasmid thus isolated, the ethidium bromide was removed by extraction with
n-butanol, and the plasmid solution was dialyzed against a TE buffer to give 100 .mu.g purified
recombinant plasmid pT1.

10. Document ID: US 5629171 A

L4: Entry 10 of 20
File: USPT
May 13, 1997

US-PAT-NO: 5629171
DOCUMENT-IDENTIFIER: US 5629171 A
TITLE: Recombinant bioprocess for the preparation of 7-amino cephalosporanic acid (7-ACA)
DATE-ISSUED: May 13, 1997

US-CL-CURRENT: 435/47; 435/183, 435/230, 435/243, 435/252.3, 435/254.11, 435/254.5, 435/49, 435/51,
536/23.1, 536/23.2, 536/23.74

APPL-NO: 8/ 439404
DATE FILED: May 11, 1995

PARENT-CASE:
This application is a division of application Ser. No. 08/250,310, filed May 27, 1994, now U.S. Pat.
No. 5,559,005, which is a continuation of application Ser. No. 07/953,492, filed Oct. 6, 1992 now
abandoned, which is a continuation-in-part of application Ser. No. 07/777,833, filed Oct. 15, 1991
now abandoned.

IN: Conder; Michael J., Rambosek; John A., McAda; Phyllis C., Reeves; Christopher D.

AB: Important intermediates for preparing cephalosporin antibiotics, 7-amino-cephalosporanic acid (7-ACA) and
7-aminodeacetylcephalosporanic acid (7-ADAC), are
prepared by a novel bioprocess in which a transformed Penicillium chrysogenum strain is
cultured in the presence of an adipate feedstock to produce adipoyl-6-APA (6-amino penicillanic
acid); followed by the in situ expression of the following genes with which the P. chrysogenum
has been transformed:, 1) an expandase gene, whose expression product converts the
adipoyl-6-APA by ring expansion to adipoyl-7-ADCA;, 2) an hydroxylase gene whose expression
product converts the 3-methyl side chain of adipoyl-7-ADCA to 3-hydroxymethyl, to give the
first product, 7-aminodeacetylcephalosporanic acid (7-ADAC); and, 3) an acetyltransferase gene
whose expression product converts the 3-hydroxymethyl side chain to the 3-acetyloxymethyl side
chain of 7-ACA. The final product, 7-ACA, is then prepared by cleavage of the adipoyl side
chain using an adipoyl acylase. The entire synthesis, accordingly, is carried out using
bioprocesses, and is efficient and economical.

L4: Entry 10 of 20
File: USPT
May 13, 1997

DOCUMENT-IDENTIFIER: US 5629171 A
TITLE: Recombinant bioprocess for the preparation of 7-amino cephalosporanic acid (7-ACA)

DEPR:
E. coli cultures containing the plasmid of interest were grown in 500 mL LB broth (20 g/l of LB
Broth Base (Gibco, Paisley, Scotland), with 15 .mu.g/mL tetracycline on a rotary shaker at 220 rpm
for 12-16 hours at 37.degree. C. The cells were pelleted by centrifugation at 4000.times.g for ten
minutes at 4.degree. C. The cell pellet was resuspended in 18 mL Glucose Buffer (50 mM glucose, 25
mM Tris pH 8.0, 10 mM EDTA) and 2 mL of 40 mg/mL lysozyme (Sigma, St. Louis, Mo.) in glucose buffer
was added, mixed, and the mixture was incubated at room temperature for 15 minutes. Forty mL of a
freshly prepared solution of 0.2N NaOH, 1% SDS was added, and the mixture swirled gently and placed
on ice for ten minutes. Thirty mL of 5M potassium acetate pH 4.8 were

then added, mixed well, and
the resultant mixture was placed on ice for an additional ten minutes. The cellular debris were
pelleted by centrifugation at 4000.times.g for ten minutes at 4.degree. C. and the resulting
supernatant was filtered through a cheesecloth filter. Isopropanol (0.6 volumes) was added to the
clarified supernatant to precipitate the plasmid DNA, and the precipitate was formed during
incubation at room temperature for 20 minutes. The plasmid DNA was pelleted at 4000.times.g for 20
minutes at 4.degree. C. and then washed with 70% ethanol and dried briefly. The pellet was
resuspended in 9 mL TE buffer, then 10 grams of CsCl and 0.387 mL of a 10 mg/mL ethidium bromide
solution were added. This solution was centrifuged at 313,100.times.g for 24 hours. The resulting
plasmid band in the cesium chloride gradient was visualized with ultraviolet light, isolated, and
then the ethidium bromide was removed using water saturated butanol for extraction. The CsCl in the
plasmid preparation was then removed by dialysis against TE buffer, and finally the DNA was
concentrated using PEG (MW 8000). Concentration of DNA was determined spectrophotometrically using
an absorbance reading at 260 nm.

11. Document ID: US 5559005 A

L4: Entry 11 of 20

File: USPT

Sep 24, 1996

US-PAT-NO: 5559005
DOCUMENT-IDENTIFIER: US 5559005 A
TITLE: Bioprocess for preparing 7-ACA and 7-ADAC
DATE-ISSUED: September 24, 1996

US-CL-CURRENT: 435/47; 435/183, 435/230, 435/243, 435/254.11, 435/254.5, 435/320.1, 435/49, 435/51,
536/23.1, 536/23.2, 536/23.74

APPL-NO: 8/ 250310
DATE FILED: May 27, 1994

PARENT-CASE:
RELATED APPLICATIONS This is a continuation of application Ser. No. 07/953,492 filed on Oct. 6, 1992
and now abandoned and which is a continuation-in-part application of application Ser. No.
07/777,833, filed on Oct. 15, 1991 and now abandoned.

IN: Conder; Michael J., McAda; Phyllis C., Rambosek; John A., Reeves; Christopher D.

AB: Important intermediates for preparing cephalosporin antibiotics, 7-amino-cephalosporanic acid (7-ACA) and 7-aminodeacetylcephalosporanic acid (7-ADAC), are
prepared by a novel bioprocess in which a transformed Penicillium chrysogenum strain is
cultured in the presence of an adipate feedstock to produce adipoyl-6-APA (6-amino penicillanic
acid); followed by the in situ expression of the following genes with which the P. chrysogenum
has been transformed:, 1) an expandase gene, e.g., from Cephalosporium acremonium, whose
expression product converts the adipoyl-6-APA by ring expansion to adipoyl-7-ADCA;, 2) an
hydroxylase gene whose expression product converts the 3-methyl side chain of adipoyl-7-ADCA to
3-hydroxymethyl, to give the first product, 7-aminodeacetylcephalosporanic acid (7-ADAC); and,
3) an acetyltransferase gene whose expression product converts the 3-hydroxymethyl side chain
to the 3-acetyloxymethyl side chain of 7-ACA. The final product, 7-ACA, is then prepared by
cleavage of the adipoyl side chain using an adipoyl acylase. The entire synthesis, accordingly,
is carried out using bioprocesses, and is efficient and economical.

L4: Entry 11 of 20

File: USPT

Sep 24, 1996

DOCUMENT-IDENTIFIER: US 5559005 A
TITLE: Bioprocess for preparing 7-ACA and 7-ADAC

DEPR:
E. coli cultures containing the plasmid of interest were grown in 500 mL LB broth (20 g/l of LB
Broth Base (Gibco, Paisley, Scotland), with 15 .mu.g/mL tetracycline on a rotary shaker at 220 rpm
for 12-16 hours at 37.degree. C. The cells were pelleted by centrifugation at 4000.times.g for ten
minutes at 4.degree. C. The cell pellet was resuspended in 18 mL Glucose Buffer (50 mM glucose, 25
mM Tris pH8.0, 10 mM EDTA) and 2 mL of 40 mg/mL lysozyme (Sigma, St. Louis, Mo.) in glucose buffer
was added, mixed, and the mixture was incubated at room temperature for 15 minutes. Forty mL of a
freshly prepared solution of 0.2N NaOH, 1% SDS was added, and the mixture swirled gently and placed
on ice for ten minutes. Thirty mL of 5M potassium acetate pH 4.8 were then added, mixed well, and
the resultant mixture was placed on ice for an additional ten minutes. The cellular debris were
pelleted by centrifugation at 4000.times.g for ten minutes at 4.degree. C. and the resulting
supernatant was filtered through a cheesecloth filter. Isopropanol (0.6 volumes) was added to the
clarified supernatant to precipitate the plasmid DNA, and the precipitate was formed during
incubation at room temperature for 20 minutes. The plasmid DNA was pelleted at 4000.times.g for 20
minutes at 4.degree. C. and then washed with 70% ethanol and dried briefly. The pellet was
resuspended in 9 mL TE buffer, then 10 grams of CsCl and 0.387 mL of a 10 mg/mL ethidium bromide
solution were added. This solution was centrifuged at 313, 100.times.g for 24 hours. The resulting
plasmid band in the cesium chloride gradient was visualized with ultraviolet light, isolated, and
then the ethidium bromide was removed using water saturated butanol for extraction. The CsCl in the
plasmid preparation was then removed by dialysis against TE buffer, and finally the DNA was
concentrated using PEG (MW 8000). Concentration of DNA was determined spectrophotometrically using
an absorbance reading at 260 nm.

12. Document ID: US 5432071 A

L4: Entry 12 of 20

File: USPT

Jul 11, 1995

US-PAT-NO: 5432071
DOCUMENT-IDENTIFIER: US 5432071 A

TITLE: Variant E1 protein gene for pyruvate dehydrogenase complex and variant E1 protein of pyruvate
dehydrogenase complex
DATE-ISSUED: July 11, 1995

US-CL-CURRENT: 435/190; 435/189, 435/252.33, 435/320.1, 435/69.1, 536/23.2

APPL-NO: 8/ 215709
DATE FILED: March 22, 1994

FOREIGN-APPL-PRIORITY-DATA:
COUNTRY APPL-NO APPL-DATE
JP 4-082130 April 8, 1993

IN: Ichikawa; Toshio, Koyama; Yasuji, Otake;Hideko, Nakano; Eiichi

AB: The present invention relates to variant E.sub.1 protein of pyruvate dehydrogenase
complex of high activity in which arginine at the 146-position is replaced by proline in the
amino acid sequence of wild-type E.sub.1 protein of pyruvate dehydrogenase complex, a gene
coding for said variant E.sub.1 protein, a novel recombinant DNA comprising said variant
E.sub.1 protein-encoding gene inserted in a vector DNA, and a process for producing variant
E.sub.1 protein by said recombinant DNA. The present invention provides the variant E.sub.1
protein of pyruvate dehydrogenase complex of high activity.

L4: Entry 12 of 20 File: USPT Jul 11, 1995

DOCUMENT-IDENTIFIER: US 5432071 A
TITLE: Variant E1 protein gene for pyruvate dehydrogenase complex and variant E1 protein of pyruvate
dehydrogenase complex

DEPR:
The precipitate was dried under reduced pressure and then dissolved in 6 ml of 10 mM Tris-HCl
buffer, pH 7.5, containing 1 mM EDTA, followed by addition of 6 g cesium chloride and 0.3 ml of 10
mg/ml ethidium bromide. For isolation of the recombinant plasmid pACEE1K' DNA, the sample solution
was subjected to equilibrium density-gradient centrifugation for 20 hours at 50000 r.p.m. by means
of a ultracentrifugate. After ethidium bromide was removed by extraction with n-butanol, the sample
solution was dialyzed against 10 mM Tris-HCl buffer (pH 7.5) containing 1 mM EDTA, to give 100 .mu.g
of purified recombinant plasmid pACEE1K' DNA.

13. Document ID: US 5318896 A

L4: Entry 13 of 20 File: USPT Jun 7, 1994

US-PAT-NO: 5318896
DOCUMENT-IDENTIFIER: US 5318896 A
TITLE: Recombinant expandase bioprocess for preparing 7-aminodesacetoxy cephalosporanic acid
(7-ADCA)
DATE-ISSUED: June 7, 1994

US-CL-CURRENT: 435/47; 435/183, 435/230, 435/51, 435/935

APPL-NO: 7/ 933469
DATE FILED: August 28, 1992

PARENT-CASE:
This application is a continuation-in-part of application Ser. No. 07/757,879, filed Sep. 11, 1991,
now abandoned.

IN: Conder; Michael J., McAda; Phyllis C., Rambosek; John A.

AB: An important intermediate for preparing cephalosporin antibiotics, 7-aminodesacetoxy
cephalosporanic acid (7-ADCA), is prepared by a novel bioprocess in which a transformed
Penicillium chrysogenum strain is cultured in the presence of an adipate feedstock to produce
adipoyl-6-APA (6-amino penicillanic acid); and the in situ expression of an expandase gene,
e.g., from Streptomyces clavuligerus, with which the P. chrysogenum has been transformed,
converts the adipoly-6-APA by ring expansion to adipoyl-7-ADCA. The final product, 7-ADCA, is
then prepared by cleavage of the adipoyl side chain using an adipoyl acylase. The entire
synthesis, accordingly, is carried out using bioprocesses, and is efficient and economical.

L4: Entry 13 of 20 File: USPT Jun 7, 1994

DOCUMENT-IDENTIFIER: US 5318896 A
TITLE: Recombinant expandase bioprocess for preparing 7-aminodesacetoxy cephalosporanic acid
(7-ADCA)

DEPR:
E. coli cultures containing the plasmid were grown in 500 mL LB broth (20 g/1 of LB Broth Base
(Gibco, Paisley, Scotland), with 15 mg/mL tetracycline on a rotary shaker at 220 rpm for 12-16 hours
at 37.degree. C. The cells were pelleted by centrifugation at 4000.times.g for ten minutes at
4.degree. C. The cell pellet was resuspended in 18 mL Glucose Buffer (50 mM glucose, 25 mM Tris
pH8.0, 10 mM EDTA) and 2 mL of 40 mg/mL lysozyme (Sigma, St. Louis, Mo.) in glucose buffer was
added, mixed, and the mixture was incubated at room temperature for 15 minutes. Forty mL of a
freshly prepared solution of 0.2N NaOH, 1% SDS was added, and the mixture swirled gently and placed
on ice for ten minutes. Thirty mL of 5M potassium acetate pH 4.8 were then added, mixed well, and
the resultant mixture was placed on ice for an additional ten minutes. The cellular debris were
pelleted by centrifugation at 400.times.g for ten minutes at 4.degree. C. and the resulting
supernatant was filtered through a cheesecloth filter. Isopropanol (0.6 volumes) was added to the
clarified supernatant to precipitate the plasmid DNA, and the precipitate was formed during
incubation at room temperature for 20 minutes. The plasmid DNA was pelleted at 4000.times.g for 20
minutes at 4.degree. C. and then washed with 70% ethanol and dried briefly. The pellet was

resuspended in 9 mL TE buffer, then 10 grams of CsCl and 0.387 mL of a 10 mg/mL ethidium bromide
solution were added. This solution was centrifuged at 313,100.times.g for 24 hours. The resulting
plasmid band in the cesium chloride gradient was visualized with ultraviolet light, isolated, and
then the ethidium bromide was removed using water saturated butanol for extraction. The CsCl in the
plasmid preparation was then removed by dialysis against TE buffer, and finally the DNA was
concentrated using PEG (MW 8000). Concentration of DNA was determined spectrophotometrically using
an absorbance reading at 260 nm.

14. Document ID: US 5175104 A

L4: Entry 14 of 20

File: USPT

Dec 29, 1992

US-PAT-NO: 5175104
DOCUMENT-IDENTIFIER: US 5175104 A
TITLE: Recombinant DNA and a process for producing phosphotransacetylase
DATE-ISSUED: December 29, 1992

US-CL-CURRENT: 435/194; 435/252.33, 435/320.1

APPL-NO: 7/ 452388
DATE FILED: December 19, 1989

FOREIGN-APPL-PRIORITY-DATA:

COUNTRY	APPL-NO	APPL-DATE
JP	63-329314	December 28, 1988

IN: Asahi; Matsuyama, Hideko; Otake, Eiichi; Nakano

AB: A novel recombinant DNA is characterized in that DNA containing a gene encoding
phosphotransacetylase has been inserted into vector DNA. The recombinant DNA can be obtained by
culturing in medium E. coli 1100 (pPT200) (FERM BP-2195) belonging to the genus Escherichia
which contains a recombinant DNA obtained by inserting DNA containing a gene coding for
phosphotransacetylase into vector DNA and is capable of producing phosphotransacetylase, and
collecting phosphotransacetylase from the culture.

L4: Entry 14 of 20

File: USPT

Dec 29, 1992

DOCUMENT-IDENTIFIER: US 5175104 A
TITLE: Recombinant DNA and a process for producing phosphotransacetylase

DEPR:
The precipitates were dried under reduced pressure in a conventional manner and dissolved in 6 ml of
10 mM Tris-hydrochloride buffer (pH 7.5) containing 1 mM EDTA. Furthermore, 6 g of cesium chloride
and 0.2 ml of 10 mg/ml ethydium bromide were added to the solution. The mixture was subjected to
equilibrium density gradient centrifugation at 39,000 r.p.m. for 42 hours in a conventional manner,
using a ultracentrifuging machine to isolate the recombinant plasmid pPT100 DNA. Further after
ethydium bromide was removed using n-butanol, dialysis was performed to 10 mM Tris-hydrochloride
buffer (pH 7.5) containing 1 mM EDTA to give 1500 .mu.g of the purified recombinant plasmid pPT100
DNA (the size was ca. 10.0 Kb).

DEPR:
The precipitates were then dried under reduced pressure in a conventional manner and dissolved in 6
ml of 10 mM Tris-hydrochloride buffer (pH 7.5) containing 1 mM EDTA. Furthermore, 6 g of cesium
chloride and 0.2 ml of ethydium bromide (19 mg/ml) were added to the solution. The mixture was
subjected to equilibrium density gradient centrifugation at 39,000 r.p.m. for 42 hours in a
conventional manner, using a ultracentrifuging machine to isolate the recombinant plasmid pPT200
DNA. Further after ethydium bromide was removed using n-butanol, dialysis was performed to 10 mM
Tris-hydrochloride buffer (pH 7.5) containing 1 mM EDTA to give 600 .mu.g of the purified
recombinant plasmid pPT200 DNA.

15. Document ID: US 4968613 A

L4: Entry 15 of 20

File: USPT

Nov 6, 1990

US-PAT-NO: 4968613
DOCUMENT-IDENTIFIER: US 4968613 A
TITLE: Luciferase gene and novel recombinant DNA as well as a method of producing luciferase
DATE-ISSUED: November 6, 1990

US-CL-CURRENT: 435/189; 435/320.1, 536/23.2, 536/23.7

APPL-NO: 7/ 224445
DATE FILED: July 26, 1988

FOREIGN-APPL-PRIORITY-DATA:

COUNTRY	APPL-NO	APPL-DATE
JP	62-187724	July 29, 1987
JP	62-187725	July 29, 1987
JP	62-205194	August 20, 1987

IN: Masuda; Tsutomu, Tatsumi; Hiroki, Nakano; Eiichi

AB: A luciferase gene isolated from Luciola cruciata (Japanese firefly) coding for an
amino acid sequence shown in FIG. 4 and a novel recombinant DNA characterized by incorporating
a gene coding for luciferase into a vector DNA are disclosed. There is also disclosed a method
of producing luciferase which comprises culturing in a medium a

microorganism containing a
recombinant DNA having inserted a gene coding for luciferase in a vector DNA and belonging to
the genus Escherichia capable of producing luciferase and collecting luciferase from the
culture.

L4: Entry 15 of 20
File: USPT
Nov 6, 1990

DOCUMENT-IDENTIFIER: US 4968613 A
TITLE: Luciferase gene and novel recombinant DNA as well as a method of producing luciferase

DEPR:
Then, the precipitates were dried under reduced pressure in a conventional manner and dissolved in
10 mM Tris-hydrochloride buffer (pH 7.5) containing 1 mM EDTA. To the solution were further added 6
g of cesium chloride and 0.2 ml of ethydium bromide solution (10 mg/ml). The resulting mixture was
subjected to an equilibrated density gradinent centrifugation treatment using a ultracentrifuging
machine at 39,000 r.p.m. for 42 hours in a conventional manner thereby to isolate recombinant
plasmid pMCE 10 DNA. After ethydium bromide was removed using n-butanol, dialysis was performed to
10 mM Tris-hydrochloride buffer (pH 7.5) containing 1 mM EDTA to 500 .mu.g,of purified recombinant
plasmid pMCE 10 DNA.

16. Document ID: US 4681931 A

L4: Entry 16 of 20
File: USPT
Jul 21, 1987

US-PAT-NO: 4681931
DOCUMENT-IDENTIFIER: US 4681931 A
TITLE: Process for the isolation and purification of alpha-interferons
DATE-ISSUED: July 21, 1987

US-CL-CURRENT: 530/351; 435/69.51

APPL-NO: 6/ 741865
DATE FILED: June 6, 1985

FOREIGN-APPL-PRIORITY-DATA:
COUNTRY APPL-NO APPL-DATE
DE 3421302 June 8, 1984

IN: Obermeier; Rainer, Salomon; Ingeborg, Ludwig; Jurgen

AB: The invention relates to a process for the isolation and purification of
.alpha.-interferons generated by plasmids from bacterial cultures which have been modified by
genetic engineering, and of .alpha.-interferons from culture supernatants of induced mammalian
cells, which comprises partition of appropriate crude substances containing interferon between
an aqueous urea solution, which contains a surfactant, and a mixture of n-butanol and glacial
acetic acid which contains water, and isolation of the purified .alpha.-interferons from the
upper phase.

L4: Entry 16 of 20
File: USPT
Jul 21, 1987

DOCUMENT-IDENTIFIER: US 4681931 A
TITLE: Process for the isolation and purification of alpha-interferons

ABPL:
The invention relates to a process for the isolation and purification of .alpha.-interferons
generated by plasmids from bacterial cultures which have been modified by genetic engineering, and
of .alpha.-interferons from culture supernatants of induced mammalian cells, which comprises
partition of appropriate crude substances containing interferon between an aqueous urea solution,
which contains a surfactant, and a mixture of n-butanol and glacial acetic acid which contains
water, and isolation of the purified .alpha.-interferons from the upper phase.

BSPR:
Thus the invention relates to a process for the isolation and purification of .alpha.-interferons
generated by plasmids from bacterial cultures which have been modified by genetic engineering, and
of .alpha.-interferons from culture supernatants from induced mammalian cells, which comprises
partition of appropriate crude substances containing interferons between an aqueous urea solution,
which contains a surfactant, and a mixture of n-butanol and glacial acetic acid which contains
water, and isolation of the purified .alpha.-interferons from the upper phase.

CLPR:
1. A process for the isolation and purification of .alpha.-interferons generated by plasmids from
bacterial cultures which have been modified by genetic engineering, and of .alpha.-interferons from
culture supernatants from induced mammalian cells, which comprises partition of crude materials
containing .alpha.-interferon between an aqueous urea solution, which contains an anionic
surfactant, and a mixture of n-butanol and glacial acetic acid which contains water, and isolation
of the purified .alpha.-interferons from the upper phase.

17. Document ID: US 4517300 A

L4: Entry 17 of 20
File: USPT
May 14, 1985

US-PAT-NO: 4517300
DOCUMENT-IDENTIFIER: US 4517300 A
TITLE: Plasmid pSAN 181
DATE-ISSUED: May 14, 1985

US-CL-CURRENT: 435/91.4; 435/320.1

APPL-NO: 6/ 475245

DATE FILED: March 14, 1983

FOREIGN-APPL-PRIORITY-DATA:
COUNTRY
APPL-NO
APPL-DATE
JP
57-41227
March 16, 1982

IN: Manome; Taichi, Ohmine; Toshinori, Okazaki; Takao, Arai; Mamoru

AB: A novel plasmid, named pSAN 181, has a molecular weight of 5.+-.0.2 megadaltons and
single cleavage sites for each of the restriction endonucleases BamHI, PstI, XhoI and BglII,
the positions of said sites being 0, about 0.85, about 1.15 and about 3.95 megadaltons from the
cleavage site of BamHI. The plasmid is extracted from mycelia of microorganisms of the species
Streptomyces fulvoviridis, especially the strain Streptomyces fulvoviridis FERM P-6279, and is
useful as a vector for genetic engineering.

L4: Entry 17 of 20
File: USPT
May 14, 1985

DOCUMENT-IDENTIFIER: US 4517300 A
TITLE: Plasmid pSAN 181

BSPR:
Separation and purification of the plasmid pSAN 181 from the mycelia of Streptomyces fulvoviridis
FERM P-6279 may be effected by known methods, for example as disclosed in the Journal of Antibiotics
33, 88-91 (1980). In this method,, the mycelia are suspended in a buffer containing
ethylenediaminetetraacetic acid (EDTA). An enzyme capable of dissolving cell walls, for example
lysozyme, and preferably a surfactant, such as sodium dodecyl sulphate, are then added to effect
lysis. The sodium dodecyl sulphate is precipitated by adding aqueous sodium chloride and removed;
ribonuclease and pronase are then added to the supernatant to liberate the DNA. This DNA is then
subjected to cesium chloride-ethidium bromide equilibrium density gradient centrifugation to
separate the circular DNA of plasmid pSAN 181 from the chromosomal DNA. The plasmid DNA thus
separated is collected by a suitable method, the ethidium bromide is removed with butanol, and then
the DNA is purified by dialysis. The purified product may be further purified by equilibrium density
gradient centrifugation, sucrose density gradient centrifugation or agarose gel electrophoresis, if
desired. Plasmid pSAN 181 can also be separated by alkali treatment and isolation as disclosed in
Anal. Biochemi. 76, 431-441 (1976).

18. Document ID: US 4460693 A

L4: Entry 18 of 20
File: USPT
Jul 17, 1984

US-PAT-NO: 4460693
DOCUMENT-IDENTIFIER: US 4460693 A
TITLE: Plasmids derived from Actinomycetes
DATE-ISSUED: July 17, 1984

US-CL-CURRENT: 435/320.1

APPL-NO: 6/ 379207
DATE FILED: May 17, 1982

FOREIGN-APPL-PRIORITY-DATA:
COUNTRY
APPL-NO
APPL-DATE
JP
56-72185
May 15, 1981

IN: Toyama; Hiromi, Nojiri; Chuhei, Shomura; Takashi, Yamada; Yujiro

AB: Essentially pure plasmids derived from Actinomycetes are described which have at
least one restriction cleavage site for specific restriction enzyme and have molecular weight
of less than 1.0.times.10.sup.7.

L4: Entry 18 of 20
File: USPT
Jul 17, 1984

DOCUMENT-IDENTIFIER: US 4460693 A
TITLE: Plasmids derived from Actinomycetes

DEPR:
Further, after removing ethidium bromide with n-butanol extraction, the plasmid DNA was dialyzed
against TE buffer (10 mM Tris HCl buffer and 1 mM EDTA, pH 8.0). Purification of the sample after
dialysis was carried out by 5-20% neutral sucrose density gradients centrifugation. Namely, using a
TEN buffer containing ethidium bromide (10 mM Tris HCl buffer solution, 20 mM EDTA, 50 mM sodium
chloride and 1 .mu.g/ml ethidium bromide, pH 8.0), a mixture of 0.5 ml of the sample and 0.05 ml of
ethidium bromide (5 mg/ml) was superposed on a 5-20% sucrose linear density gradients, and
centrifugal separation was carried out at 4.degree. C. for 3 hours at 40,000 r.p.m. on a Beckman
model L2-75B ultracentrifuge in a SW 40 rotor. This centrifuge tube was irradiated with 365 nm
ultraviolet, and a plasmid band emitting fluorescence was taken out. After removing ethidium bromide
with n-butanol extraction, the pSF588 plasmid DNA was dialyzed against a TE buffer to obtain a
purified plasmid.

DEPR:
Since a cell of Streptomyces griseochromogens SF 701 comprised two plasmids having each a different
molecular weight, preparation was carried out by the same procedure as in Example 1 (except that the
concentration of glycine in medium was 0.5%) by CsCl-ethidium bromide boyant density gradient
centrifugation, and then separation was carried out by 5-20% neutral sucrose density gradient
centrifugation. The centrifuge tube was irradiated with 365 nm ultraviolet, and a plasmid pSF 701-1
having a lower molecular weight which appeared the upper part of the centrifuge tube was taken out.
After removing ethidium bromide with n-butanol extraction, the plasmid DNA was dialyzed against TE
buffer to obtain purified plasmid pSF 701-1.

19. Document ID: US 4681931 A

L4: Entry 19 of 20
File: EPAB
Jul 21, 1987

PUB-NO: US004681931A
DOCUMENT-IDENTIFIER: US 4681931 A
TITLE: Process for the isolation and purification of alpha-interferons

PUBN-DATE: July 21, 1987

INT-CL (IPC): C07K 14/56
EUR-CL (EPC): C07K014/56

APPL-NO: US74186585
APPL-DATE: June 6, 1985

PRIORITY-DATA: DE03421302A (June 8, 1984)

IN: OBERMEIER, RAINER, SALOMON, INGEBORG, LUDWIG, JUERGEN

AB: CHG DATE=19990617 STATUS=O>The invention relates to a process for the isolation and purification of alpha -interferons generated by plasmids from bacterial cultures which have been modified by genetic engineering, and of alpha -interferons from culture supernatants of induced mammalian cells, which comprises partition of appropriate crude substances containing interferon between an aqueous urea solution, which contains a surfactant, and a mixture of n-butanol and glacial acetic acid which contains water, and isolation of the purified alpha -interferons from the upper phase.

L4: Entry 19 of 20
File: EPAB
Jul 21, 1987

DOCUMENT-IDENTIFIER: US 4681931 A
TITLE: Process for the isolation and purification of alpha-interferons

FPAR:
CHG DATE=19990617 STATUS=O>The invention relates to a process for the isolation and purification of alpha -interferons generated by plasmids from bacterial cultures which have been modified by genetic engineering, and of alpha -interferons from culture supernatants of induced mammalian cells, which comprises partition of appropriate crude substances containing interferon between an aqueous urea solution, which contains a surfactant, and a mixture of n-butanol and glacial acetic acid which contains water, and isolation of the purified alpha -interferons from the upper phase.

20. Document ID: EP 164069 A, AU 8543422 A, DE 3421302 A, DE 3569123 G, DK 8502544 A, EP 164069 B, ES 8604022 A, FI 8502251 A, JP 61009298 A, NO 8502313 A, PT 80617 A, US 4681931 A, ZA 8504329 A

L4: Entry 20 of 20
File: DWPI
Dec 11, 1985

DERWENT-ACC-NO: 1985-311996
DERWENT-WEEK: 198550

TITLE: Isolating and purifying alpha interferon - from genetically modified bacteria or animal cells, by partitioning between aq. urea and organic phase

PRIORITY-DATA: 1984DE-3421302 (June 8, 1984)

PATENT-FAMILY:

PUB-NO	PUB-DATE	LANGUAGE	PAGES	MAIN-IPC
EP 164069 A	December 11, 1985	G	008	
AU 8543422 A	December 12, 1985		000	
DE 3421302 A	December 12, 1985		000	
DE 3569123 G	May 3, 1989		000	
DK 8502544 A	December 9, 1985		000	
EP 164069 B	March 29, 1989	E	000	
ES 8604022 A	June 1, 1986		000	
FI 8502251 A	December 9, 1985		000	
JP 61009298 A	January 16, 1986		000	
NO 8502313 A	December 30, 1985		000	
PT 80617 A	March 17, 1986		000	
US 4681931 A	July 21, 1987		000	
ZA 8504329 A	December 11, 1985		000	

APPLICATION-DATA:

PUB-NO	APPL-DATE	APPL-NO	DESCRIPTOR
EP 164069A	May 30, 1985	1985EP-0106648	
ES 8604022A	June 5, 1985		

1985ES-0543916

JP61009298A
June 7, 1985
1985JP-0122856

US 4681931A
June 6, 1985
1985US-0741865

ZA 8504329A
June 7, 1985
1985ZA-0004329

INT-CL (IPC): A61K 39/39; A61K 45/02; C07K 3/12; C07K 15/26; C12N 15/00; C12P 21/02

IN: LUDWIG, J, OBERMEIER, R, SALOMON, I

AB: Plasmid-derived alpha-interferons (I) from genetically-modified bacterial cultures
or (I) from culture supernatants of induced animal cells are isolated and purified by
partitioning the crude material between (1) a surfactant-contg. aq urea soln and (2) a
water-contg. n-butanol/ acetic acid mixt. The purified (I) is then isolated from the upper
phase., USE/ADVANTAGE - The method is esp. used to recover (I) from genetically-en gineered
E.coli cells. It eliminates 95% of the impurities from the fermentation prod in a single stage
and is simpler than known affinity or adsorption procedures., A process for the isolation and
purification of alpha-interferons generated by plasmids from bacterial cultures which have been
modified by genetic engineering, and from culture supernatants from induced mammalian cells,
which comprises partition of appropriate crude substances containing interferon between an
aqueous urea solution, which contains a surfactant, and a mixture of n-butanol and glacial
acetic acid which contains water, and isolation of the purified alpha-interferons from the
upper phase., (4pp), Isolation and purification of alpha-interferons comprises partition of
crude materials between an aq. urea soln. contg. an anionic surfactant, and a mixt. of
n-butanol and glacial acetic acid contg. water, followed by isolation of the purified prod.
from the upper phase. The alpha-interferons are generated by plasmids from bacterial cultures
which have been modified by genetic engineering, or are obtd. from culture supernatants of
induced mammalian cells., Pref. the lower phase contains 1-8 mol/l urea and 0.1-20% sodium
dodecyl sulphate. Pref. upper phase is a mixt. of n-butanol/ glacial acetic acid/water in ratio
by vol. of 215:175:2150., ADVANTAGE - Method enables elimination of more than 95% of all
concomitant contaminants from E.coli fermentation, which is used for large scale industrial
prodn. of alpha-interferons, for use as antivirals. (3pp)e

L4: Entry 20 of 20

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DERWENT-ACC-NO: 1985-311996
DERWENT-WEEK: 198550

TITLE: Isolating and purifying alpha interferon - from genetically modified bacteria or animal
cells, by partitioning between aq. urea and organic phase

ABEQ:
A process for the isolation and purification of alpha-interferons generated by plasmids from
bacterial cultures which have been modified by genetic engineering, and from culture supernatants
from induced mammalian cells, which comprises partition of appropriate crude substances containing
interferon between an aqueous urea solution, which contains a surfactant, and a mixture of n-butanol
and glacial acetic acid which contains water, and isolation of the purified alpha-interferons from
the upper phase.

L10 ANSWER 29 OF 47 HCAPLUS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 1992:3208 HCAPLUS
DOCUMENT NUMBER: 116:3208
TITLE: ***Separation*** of nucleic acids from biological samples with alcohols and protein denaturants
INVENTOR(S): Kamata, Kazuya; Satsuka, Toshiaki
PATENT ASSIGNEE(S): Tosoh Corp., Japan
SOURCE: Jpn. Kokai Tokkyo Koho, 6 pp.
CODEN: JKXXAF
DOCUMENT TYPE: Patent
LANGUAGE: Japanese
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
JP 03101688	A2	19910426	JP 1989-237327	19890914

AB Nucleic acids can be sepd. from samples (or fermn. cultures) by treating the sample with a protein denaturant (e.g. guanidine salt, urea, etc.) and then with .gtoreq.1 alc. (selected from e.g. EtOH, PrOH, BuOH, pentanol and hexanol) for pptn. or removal. No harmful solvent is useful in the extn. Thus, ***plasmid*** pIBI176-transformed Escherichia coli culture medium was treated with guanidine-HCl and stirred with iso-PrOH. After centrifugation, the pptn. was washed with 70% EtOH twice, dissolved in pH 8.0 Tris buffer contg. 1 mM EDTA and again centrifuged. The resulting supernatant was treated with EtOH and centrifuged to give DNA.